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Multiscale modelling of blood flow and transport in the human placenta

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Supervised by
Dr Alys R Clark
Professor Merryn H Tawhai

A thesis submitted in fulfilment of the requirements for the degree of Doctor of Philosophy in Bioengineering.

Auckland Bioengineering Institute
The University of Auckland

March 2017
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The undersigned hereby certify that:

- the above statement correctly reflects the nature and extent of the PhD candidate’s contribution to this work, and the nature of the contribution of each of the co-authors; and

- that the candidate wrote all or the majority of the text.

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Abstract

The placenta is critical to fetal survival and health. To prevent and/or manage complicated pregnancies, it is desirable to understand how placental structures influence its function. In this thesis, a novel modelling framework was developed to simulate blood flow and transport in the human placenta. The model includes the well-defined branching structure of the villous tree branches, as well as a smoothed representation of the terminal villi that comprise the placenta’s gas exchange interfaces. Inclusion of these structural characteristics, which are missing from previous models, has provided a new approach for investigating the structure-function relationship of the placenta.

The framework was first applied to model blood flow in a functional unit (i.e. villous tree). Studies using this model have demonstrated the sensitivity of maternal blood flow in the intervillous space (IVS) to the branching geometry of the villous tree. Distribution of villous branches was found to influence the depth of penetration by maternal blood into the IVS. When the villous branches are spaced sufficiently apart, the model predicted that maternal blood bypasses villous tissue through shunts between spiral arteries (SAs) and decidual veins (DVs), which is detrimental to material exchange in the placenta.

The blood flow model was extended to include nuclear magnetic resonance (NMR) signal generation and parameter estimation using the intravoxel incoherent motion (IVIM) method. The model was used to predict and relate IVIM parameters with the structural and blood flow properties of the placenta. Findings from this model suggest that the conventional meanings of IVIM parameters, which were previously derived for tissues with a single circulatory system, are not directly applicable to placental images as the placenta contains two independent maternal and fetal blood flow systems. The model demonstrates that placental IVIM parameters are lumped parameters representative of both blood flow systems and hence should be considered carefully when interpreting placental images.

The blood flow model was then coupled to a model of oxygen transport to investigate how structural heterogeneity of the placenta affects oxygen transport and uptake in normal and pathological pregnancies. To be an efficient exchanger, the model indicates that villous tree must maximise the exchange surface area while minimising its obstruction to maternal blood flow in the IVS. This work also provides insight into the sensitivity of oxygen exchange efficiency to villous tree geometry, such as the villous branch length and volume, which are known to change in pathologies such as intrauterine growth restriction (IUGR).

To better represent the structure of a placenta, the blood flow and oxygen transport model was upscaled from a functional unit to an organ level. The organ level model revealed that blood flow and oxygen transport are not confined within a placental functional unit, and provides motivation to shift future models towards the scale of organ
level. The modelling framework presented here not only better reflects the anatomy of the placenta but also provides a readily customisable infrastructure for application to different research questions in placental physiology and imaging.
Acknowledgements

I would like to take this opportunity to express my gratitude to everyone who contributed to this research in one way or another.

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Nothing makes me happier at work than the wonderful and supportive friends I have made here at ABI, so big shout-outs to Josh, Karthik, Jess, Vicky, Alex, Wendy, Nancy, Prasad, Naz, Mahyar, Rojan, Win, Izza, and the list goes on. Thank you for always brightening up my day with so much joy and laughter!

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List of Abbreviations

BOLD  Blood Oxygen Level Dependent imaging
CC    Central Cavity
DV    Decidual Vein
IUGR  Intrauterine Growth Restriction
IVIM  Intravoxel Incoherent Motion
IVS   Intervillous Space
MFEM  Mixed Finite Element Method
Micro-CT Micro-Computed Tomography
MRI   Magnetic Resonance Imaging
NMR   Nuclear Magnetic Resonance
PGSE  Pulsed Gradient Spin Echo
pO$_2$  Partial pressure of Oxygen
RF    Radiofrequency
RT0   Zero order Raviart-Thomas
SA    Spiral Artery
TB    Terminal tissue Block
# Chapter 3

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<th>Symbol</th>
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<tr>
<td>( n_{SA} )</td>
<td>Number of SAs in domain</td>
</tr>
<tr>
<td>( n_{DV} )</td>
<td>Number of DVs in domain</td>
</tr>
<tr>
<td>( n_{villous} )</td>
<td>Number of villous trees</td>
</tr>
<tr>
<td>( l_s )</td>
<td>Length of villous stem</td>
</tr>
<tr>
<td>( d_s )</td>
<td>Diameter of villous stem</td>
</tr>
<tr>
<td>( n_b )</td>
<td>Number of branching generations</td>
</tr>
<tr>
<td>( l_d/l_p )</td>
<td>Daughter to parent branch length ratio</td>
</tr>
<tr>
<td>( d_d/d_p )</td>
<td>Daughter to parent branch diameter ratio</td>
</tr>
<tr>
<td>( \theta_b )</td>
<td>Branch angle</td>
</tr>
<tr>
<td>( n_{seed} )</td>
<td>Number of seed points</td>
</tr>
<tr>
<td>( l_{min} )</td>
<td>Minimum length of villous branch</td>
</tr>
<tr>
<td>( \theta_{max} )</td>
<td>Maximum branch angle of daughter branch from its parent branch</td>
</tr>
<tr>
<td>( f_{dist} )</td>
<td>Fractional distance that each branch grows to the centre of mass</td>
</tr>
<tr>
<td>( \phi_{TV} )</td>
<td>Tissue density of terminal villi</td>
</tr>
<tr>
<td>( d_{TV} )</td>
<td>Average diameter of terminal villi</td>
</tr>
<tr>
<td>( n_{TV} )</td>
<td>Number of terminal convolutes branching from an intermediate villus</td>
</tr>
<tr>
<td>( l_{TV} )</td>
<td>Cumulative length of a terminal convolute</td>
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<tr>
<td>( \kappa )</td>
<td>Hydraulic conductivity</td>
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<td>( V )</td>
<td>Blood velocity in the IVS</td>
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<td>( \mu )</td>
<td>Blood viscosity</td>
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</tr>
<tr>
<td>( P_{out} )</td>
<td>Maternal blood pressure at DV</td>
</tr>
<tr>
<td>( Q_{in} )</td>
<td>Volumetric flow from an SA</td>
</tr>
<tr>
<td>( \epsilon )</td>
<td>Domain width</td>
</tr>
<tr>
<td>( d_{villi} )</td>
<td>Average diameter of villi</td>
</tr>
</tbody>
</table>
LIST OF SYMBOLS

\[ \phi \] Tissue density of villi
\[ \kappa_{\text{empty}} \] Maximum hydraulic conductivity
\[ \tau \] Domain thickness
\[ x_v \] Distance between SA and DV
\[ d \] Diameter of SA and DVs

**Chapter 4**

\[ D \] Diffusion coefficient of water in stationary tissue
\[ D^* \] Apparent diffusion coefficient of water in moving blood
\[ I \] Spin quantum number
\[ I \] Total intrinsic angular momentum of the nucleus
\[ h \] Planck’s constant
\[ \mu_n \] Magnetic dipole moment of the nucleus
\[ \gamma \] Gyromagnetic ratio of the nucleus
\[ q_n \] Nucleus charge
\[ m_n \] Nucleus mass
\[ g_f \] Dimensionless magnetic moment of the nucleus (g-factor)
\[ B \] Magnetic field
\[ \tau_m \] Torque of the nucleus
\[ \omega_L \] Larmor frequency of spin
\[ \mathbf{M} \] Net magnetisation of the magnetic moment
\[ M_x, M_y, M_z \] Scalar components of net magnetisation
\[ \hat{i}, \hat{j}, \hat{k} \] Unit vectors in direction of the \( x \)-, \( y \)-, and \( z \)-axis
\[ t \] Time
\[ M_0 \] Net magnetisation at equilibrium or at the start of PGSE sequence
\[ T_2 \] T2 relaxation rate
\[ T_1 \] T1 recovery rate
\[ \delta \] Duration of gradient pulse
\[ G \] Magnitude of gradient pulse
\[ t_1 \] Time first gradient pulse is applied in PGSE
\[ \Delta \] Time between application of first and second gradient pulse
\[ TE \] PGSE echo time
\[ B_0 \] Magnetic field at the start of PGSE sequence
$S$ \quad PGSE signal amplitude

$v, \mathbf{v}$ \quad Blood flow velocity

$m$ \quad Transverse magnetisation

$\omega_0$ \quad Larmor frequency of spin in magnetic field $B_0$

$\psi$ \quad Magnetisation that is unaffected by T2 relaxation

$A$ \quad Magnitude of $\psi$

$\varphi$ \quad Phase of $\psi$

$V$ \quad Voxel volume

$b$ \quad Diffusion sensitivity parameter

$f$ \quad Fraction of moving blood in an image pixel

$\theta$ \quad Angle between longitudinal axis of vessel and the $z$-axis

$\sigma$ \quad Angle of rotation of vessel about $x$-$y$ plane

$r$ \quad Radius of vessel segment

$l$ \quad Length of vessel segment

$d_A$ \quad Diameter of villus artery in villous stem

$D_{SR}$ \quad Strahler diameter ratio

$d_v$ \quad Diameter of fetal venous vessel

$\kappa_{uniform}$ \quad Average hydraulic conductivity of domain

$R_v$ \quad Flow resistance in fetal vessel segment

$\Delta P_v$ \quad Pressure drop across vessel segment

$Q_v$ \quad Volumetric blood flow through vessel segment

$Q_p$ \quad Volumetric blood flow through parent vessel

$Q_{d1}, Q_{d2}$ \quad Volumetric blood flow through daughter vessels

$Q_{in}$ \quad Volumetric blood flow through fetal blood flow inlet

$P_t$ \quad Blood pressure at terminal vessels

$\varphi^*$ \quad Phase of magnetisation before $\pi$ rad RF pulse

$b_N$ \quad Number of branches in each Strahler order

$< l >$ \quad Average vessel length in vessel network

$< v >$ \quad Average flow velocity in vessel network

$D_p$ \quad Pseudo-diffusion coefficient
**LIST OF SYMBOLS**

**Chapter 5**

$C_m$ Oxygen concentration in maternal blood

$D_{O_2}$ Diffusivity of oxygen in maternal blood

$C_f$ Oxygen concentration in the fetal blood bloodstream

$\alpha$ Oxygen uptake constant

$C_{in}$ Oxygen concentration in maternal blood entering through SA

$n$ Outward facing normal to the boundary

$\tau_{vm}$ Average harmonic thickness of the villous membranes

**Chapter 6**

$\eta$ Domain length

**Appendix A**

$K$ Proportionality constant in Darcy’s Equation

$\nu_E$ Scalar weighting function

$\Omega$ Domain

$\omega$ Vector weighting function

$\Gamma$ Domain boundary

$\text{Ne}$ Total number of elements in the domain

$E$ Element number

$P_E$ Pressure of element E

$\text{Ned}$ Total number of element edges (2D) or faces (3D) in the domain

$Q_i$ Normal component of velocity across edge or surface $i$

$\omega_i$ RT0 shape function for global edge (2D) or surface (3D) $i$

$P_1, P_2, P_3$ Vertices of a 2D triangular RT0 element

$|\text{El}_i|$ Length of edge $i$

$A_\Delta$ Area of a 2D triangular RT0 element

$VX_i$ Coordinate of vertex $i$ of a 2D triangular RT0 element

$\Delta x, \Delta y, \Delta z$ Length of element in the $x$-, $y$-, and $z$-directions

$\text{ne}$ Number of edges (2D) or surfaces (3D) in an RT0 element

$Q_k$ Normal velocity across local edge (2D) or surface (3D) $k$

$\omega_k$ RT0 shape function for local edge (2D) or surface (3D) $k$

$TP_k$ Normal pressure flux on the local edge (2D) or surface (3D) $k$
LIST OF SYMBOLS

\begin{itemize}
  \item $b_D$ Dirichlet pressure boundary conditions
  \item $Q$ Vector representation of fluxes across edges (2D) or surfaces (3D)
  \item $P$ Vector representation of element pressures
  \item $\hat{u}$ Velocity field within an RT0 element
  \item $\hat{u}_i$ Normal velocity associated to edge (2D) or face (3D) $i$
  \item $\bar{x}$ Line
  \item $S(\text{high})$ $y$-intercept of exponential fit to the signal when $b > 10 \text{ s/mm}^2$
\end{itemize}

\textbf{Appendix B}

$C$ Variable of interest ($\psi$ or $C_m$)
$D_C$ Diffusion coefficient of variable $C$
$f(C)$ Sink or source as a function of $C$
$C^*$ Value of $C$ advected from its departure point
$C^{n+1}$ $C$ at the end of a timestep
$\Delta C$ Change in $C$ in a timestep arising from diffusion and uptake
$\Delta t$ Timestep
$z^*$ Departure location of a particle at the beginning of a timestep
$dt$ Size of timestep
$\varphi_r$ Relative phase shift due to advection of spin particle
$\varphi^*$ Phase at the end of a timestep $t_{k+1}$
node Total number of nodes
$C_i$ Value of $C$ at node $i$
$\xi_i$ Linear basis function associated with node $i$
nn Number of nodes in an element

\textbf{Appendix C}

$C_{\text{plasma}}$ Concentration of oxygen dissolved in maternal plasma
$S_{\text{Hb}}$ Haemoglobin saturation in blood
$k_1, k_2, k_3$ Fitted constants in modified Hill equation
$C_{\text{Hb}}$ Concentration of oxygen carried by haemoglobin
Appendix E

$C_0$  Reference oxygen concentration entering through SA

$q_0$  Reference SA inlet blood flow

$\alpha_0$  Reference oxygen uptake constant

$Da$  Damköhler number

$h_{DV}$  Position of DVs relative to placentone boundary

$L$  Radius of the placentone in Chernyavsky et al

$N_a/q_0C_0$  Absolute net uptake rate

$N_T$  Net relative uptake rate
Chapter 1

Introduction

Every year, about 3 million babies are stillborn globally, while another 4 million die in the first 4 weeks of their life [1]. With this alarming death rate (800 babies every hour), there is a pressing need to better understand the physiology of normal and pathological pregnancies so that obstetric abnormalities can be recognised and managed in a timely manner to prevent adverse fetal and neonatal outcomes [2]. Many causes of such deaths - for example preterm birth, placental abruption, asphyxia, and uteroplacental insufficiency - can be traced to the placenta [3–10].

The placenta is a key organ that develops only during pregnancy and becomes obsolete after pregnancy. During the nine months of human pregnancy, the placenta performs the roles of numerous organs. It is effectively the fetus’s lungs since it acts as an interface for exchange of respiratory gases between the maternal and fetal blood. It is the fetus’s digestive system as it transports nutrients to the fetal bloodstream. In addition, the placenta takes on the role of the kidneys as it is involved in the removal of waste materials such as urea, uric acid and creatine from fetal blood. Apart from transport and metabolic functions, the placenta also produces hormones that are essential during pregnancy [11, 12].

As the placenta is critical for supporting normal growth and development of the fetus, aberrant development of the placenta is expected to affect fetal development. Placental abnormalities are associated with a host of pregnancy complications such as pre-eclampsia [13–17], intrauterine growth restriction (IUGR) [13, 18–21], fetal maldevelopment [22–26] and chorioamnionitis [27, 28]. To date, it is unknown whether the occurrence of a certain morphological abnormality in the placenta is a root cause or a
consequence of a clinical condition, and the links between morphology and placental function remain largely hypothetical [29–32].

Unlike other organs, the placenta displays profound structural variability amongst species [12, 33]. Thus, studies based on animal models may have limited applicability for extrapolation to the human [33]. Assessment of the human placenta in vivo is challenging as techniques employed to assess function must be non-invasive and cannot compromise the pregnancy in any manner. Conventional ultrasound scans typically assess the general appearance and location of the placenta without any detailed assessment of placental function [34]. While the advent of power Doppler ultrasound and magnetic resonance imaging (MRI) have allowed visualisation of placental perfusion [35–38], the generated images are generally low in resolution. High resolution images of placental structures are typically obtained through microscopic and histological studies of delivered placentas [39–43], but it can be difficult to relate the structures derived from post-partum placentas to in utero placentas with the loss of placental turgidity after delivery, and data is almost always available only at term.

Our current knowledge is mostly derived from studying placentas at discrete time points in pregnancy, for example, towards the end of the first trimester when medical termination usually occurs, during 18–22 weeks when routine ultrasound scans are recommended, or at term after the placenta is delivered. Hence, there are also gaps in our understanding of what occurs in the placenta between these time points. It is therefore desirable to piece together all of these available data to make sense of the placental structure-function relationship throughout gestation, to bridge the current gaps in our knowledge. However, few experimental tools exist to do this.

In many organs, multiscale computational models (virtual representations of organs that bridge the gap between micro- and macroscopic data) have proven useful for constructing a consolidated picture of how discrete observations (both in vivo and in vitro) relate. These models have advanced our knowledge of physiology as well as refining diagnostics and treatment strategies [44–47]. The placenta is regulated through integration of factors across multiple spatial and temporal scales, and as such, multiscale computational models are also more likely to provide greater insights than can be achieved by considering each measurement scale independently. Furthermore, with a validated computational model, perturbations can easily be introduced into an analysis and used for predicting outcomes.
1.1. Research aims

that may not be intuitively obvious. These outcomes can then be experimentally verified and used for advancing the current knowledge of the human placenta.

Current computational models of the human placenta have either focused on modelling placental structure and function at a specific spatial scale \([48–52]\), or have adopted simplified homogeneous geometric representations of the placenta \([53–57]\). Establishing a computational model of the human placenta that spans across spatial scales and incorporates structural heterogeneity, would allow study of the interplay between factors affecting the structure-function relationship of the human placenta and the resulting impact on fetal development.

This thesis focuses on the development of a multiscale model of blood flow and material transport (for example, of oxygen) in the human placenta. This computational model is useful for: (i) elucidating aspects of normal physiology, in particular, events that are happening in the placenta which cannot be observed directly through imaging or experimental measurements; (ii) providing a predictive tool that enables hypotheses to be tested \textit{in silico} without conducting unethical or infeasible experiments on living human subjects; (iii) performing simulations with controlled perturbations to identify the key parameters and to quantify the degree of change required for these parameters to have a potential impact on placental function.

**1.1 Research aims**

The aims of this research are:

1. To develop a computational model of maternal blood flow in the placenta, which incorporates the structural characteristics of the villous tree, for investigation of how maternal blood flow is influenced by villous tree geometry.

2. To incorporate fetal blood flow and apply the placental blood flow model to predict Pulsed Gradient Spin Echo (PGSE) signal generation, and relate resulting Intravoxel Incoherent Motion (IVIM) parameters to the different flow patterns in the placenta.

3. To couple oxygen transport to maternal blood flow to identify the sensitivity of oxygen exchange function to placental structure and implications in pregnancy complications.
4. To establish an anatomically realistic organ-based modelling framework that is readily customisable with structural data for parameterisation to different gestational ages.

1.2 Thesis overview

This thesis is divided into the following chapters:

**Chapter 2** illustrates the anatomy of the human placenta and provides the background for understanding the different aspects of placental physiology which are modelled in subsequent chapters. The chapter also summarises the existing computational modelling efforts that are aimed at addressing the structure-function relationship of the placenta.

**Chapter 3** presents a maternal blood flow model for a placental subunit that can be parameterised to any stage in pregnancy. The chapter details the derivation of a non-uniform porosity field representative of the intervillous space (IVS) from a villous tree defined by a branching structure for its largest villous tree branches, and a smoothed tissue block representation for its terminal villi. The chapter also introduces numerical methods used for simulating maternal blood flow in the derived non-uniform porous medium. The effects of villous tree geometry on maternal blood flow pattern in a term placental subunit are analysed in this chapter.

**Chapter 4** demonstrates an application of the placental blood flow model to interpret IVIM parameters for blood flow in the placenta. Using a week 20 model, the fetal and maternal blood flow are considered separately to tease out how IVIM parameters vary with their respective properties before combining the two independent flow systems to evaluate how IVIM parameters relate to the flow patterns in the placenta.

**Chapter 5** investigates how oxygen distribution and oxygen uptake are influenced by the villous tree. Building on the maternal blood flow model in Chapter 3, oxygen exchange is coupled to the transport dynamics of maternal blood flow in this chapter. Further to examining how oxygen uptake is influenced by a non-uniform porosity distribution over the villous tree structure, the chapter examines how regional variations in oxygen uptake capacity affect placental efficiency.
Chapter 6 presents a modelling framework towards a closer representation of placental anatomy on an organ level. This chapter extends the maternal blood flow and oxygen exchange models to include multiple villous tree structures which are supplied by a spiral artery (SA) distribution obtained from a placental bed specimen. The model predicts maternal blood flow and oxygen exchange dynamics within a section of the placenta. The chapter highlights the need for a paradigm shift towards modelling placenta on an organ level.

Chapter 7 summarises the key findings of this thesis and presents research directions for future work.

1.3 Research contributions

This research builds on established techniques for accurately modelling the branching geometry of the villous tree, and on previous placental models for simulating blood flow and transport in the IVS as a porous medium. It includes the following original developments:

- A novel technique for representing a villous tree with a well-defined branching structure of the villous tree branches, and a smoothed representation of the terminal villi convolutes which enables derivation of an IVS conductivity field that varies spatially with villous tree properties.

- An improved approach to model maternal blood flow and oxygen exchange in an IVS that is represented by a non-uniform porous medium which accounts for the architecture of the villous tree.

- Identification of key associations between placental function and villous tree geometry, and a new mechanism for arteriovenous shunts in the placenta.

- An interpretative tool for predicting PGSE signals using a placental blood flow model, and evaluation of IVIM parameters in relation to blood flow properties in the placenta.

- The first organ-level model of a placental section with multiple villous trees supplied by an SA distribution from a placental bed specimen, and its application to investigate blood flow and oxygen transport on an organ level.
Chapter 1. Introduction

1.4 Research outputs

Journal articles


Conference proceedings


Conference presentations


Chapter 2

Background

In order to construct a multiscale computational model of the placenta, knowledge of its anatomy and function are required. This chapter summarises available data describing the structure, function and pathology of the human placenta, as well as existing computational models that aim to represent either this structure or function.

2.1 Basic anatomy of the human placenta

Figures 2.1a and b show a freshly delivered term human placenta. A normal mature human placenta is typically disc-shaped [12, 58] with an average diameter of 220 mm [59]. The delivered placenta is thicker in the central region (approximately 25 mm) and thinner in the peripheral region [59]. The placenta is connected to the fetus through the umbilical cord, which is helical in shape [12] and contains two umbilical arteries and an umbilical vein. The umbilical vein transports oxygen and nutrient-rich blood from the placenta to the fetus, whereas the umbilical arteries carry deoxygenated and nutrient-depleted blood from the fetus to the placenta. On one end, the umbilical cord is attached to the abdominal region of the fetal body and the umbilical blood vessels that are embedded in the umbilical cord form a connection with the circulatory system within the fetal body, while the other end of the umbilical cord is connected to the placenta. The placental surface connected to the umbilical cord is referred to as the fetal side or the chorionic plate (Figure 2.1a). The opposing surface of the placenta is referred to as the maternal side or the basal plate (Figure 2.1b).
Figure 2.1: A freshly delivered term human placenta and its *in utero* orientation. (a) The umbilical cord is inserted on the fetal side of the placenta. The chorionic arteries (white due to injection of milk after delivery) and chorionic veins (dark) are visible on the chorionic plate. (b) The maternal side of the placenta is divided into placental lobules of varying size. Figures obtained from Benirschke et al. [12].

Figure 2.2 provides a schematic of the human placenta. The fetal blood vessels in the placenta can be split into two categories: chorionic vessels that cover the chorionic plate, and villous vessels that branch out from the chorionic plate and are housed in branching tree structures called the placental villi.

Chorionic vessels
At the site of cord insertion into the placenta, the umbilical arteries and vein (first order vessels) branch in a dichotomous manner into second, third and sometimes fourth order vessels [60]. As these vessels branch over the chorionic plate, they form a star-like pattern centrifugal from the cord insertion site (Figure 2.1a). Some branches of these chorionic vessels perforate the chorionic plate and form the first order vessels of the villous trees. Typically, one branch will perforate the chorionic plate, while the other branch will continue to extend over the chorionic plate and divide again into higher order chorionic vessels and a first order villus vessel.

Villous vessels
As illustrated in Figure 2.2, a first order villus artery is accompanied by a first order villus vein in most cases [60]. As each of these first order villus arteries and their accompanying veins extend towards the basal plate, they follow the course of the villous tree branches and divide into smaller branches. The villus artery eventually connects with the villus vein through a tortuous capillary network in the convoluted structures at the terminals of the villous tree. The capillary network consists of numerous
2.1. Basic anatomy of the human placenta

Figure 2.2: Schematic representation of a human placenta. The placenta contains numerous chorionic villous trees which stem from the fetal side of the placenta into the intervillous space (IVS). Maternal blood enters from the spiral arteries (SAs) on the maternal side of the placenta, percolates through the villous tree and IVS before draining through the decidual veins (DVs). The fetal circulation resides in the villous trees, and generally runs along the branches in stem and intermediate villi before reaching a dilated and convoluted capillary structure in the terminal villi. Oxygen and nutrients are transferred from the maternal blood surrounding the villous tree into the fetal bloodstream across the trophoblast bilayer.

parallel capillary connections and is the typical site for material exchange given its close proximity to maternal blood [12].

The space between villous tree branches and adjacent villous trees, known as the intervillous space (IVS), is perfused with blood from the maternal bloodstream. As shown in Figure 2.2, maternal blood (carrying oxygen) enters into the placenta through spiral arteries (SAs) on the basal plate and circulates around the villous trees before returning to the main maternal blood circulatory system through the decidual veins (DVs). The villous trees are therefore often described as being ‘bathed’ in maternal
blood. With two independent circulatory systems in the placenta, the developing fetus is able to obtain oxygen from its mother without having its bloodstream coming into direct contact with the maternal bloodstream.

The ‘functional subunit’ of the placenta (also known as a placentome) is typically assumed to comprise a single villous tree, which is fed by SAs and DVs [61]. The structure of the placental villous tree, and how it evolves in pregnancy are described below in detail.

2.2 The placental villous tree

2.2.1 Morphological features of villous trees

A villous tree comprises of a thick villous trunk which stems from the chorionic plate, and branches repeatedly towards the basal plate to produce seven to 35 generations (average 15) of dichotomous branching [12, 62–64]. Each villous tree comprises a trophoblast bilayer, which is made up of syncytiotrophoblast and cytotrophoblast cells. Syncytiotrophoblast is a continuous, uninterrupted multinucleated layer which forms the outer covering of the villous tree, while cytotrophoblast cells underneath the syncytiotrophoblast layer are proliferative cells that fuse into the overlying syncytiotrophoblast to support its growth and regeneration. The trophoblast bilayer forms a structural barrier for active and passive nutrient and oxygen exchange between the maternal and fetal bloodstreams during pregnancy. Although these components can be found at any part of a villous tree, the proportion and characteristics of each component vary at different subsections of the villous tree according to their villus classification, as discussed in the next section.

2.2.2 Classification of villi

Stem villi

Stem villi make up the first 15 generations of branching in the villous tree [12, 62–64]. The calibre of stem villi decrease as the villous tree branches, with their diameters ranging from 50 $\mu$m to 3000 $\mu$m [39, 62, 63, 65, 66]. As stem villi serve primarily as mechanical support to the villous tree, stem villi are characterised by a uniformly thick trophoblast bilayer, a compact fibrous stroma, and a centrally located artery and vein [12, 62]. They
also act as a delivery system to channel fetal blood to the sites for material exchange without active participation in material exchange across their surfaces [65–67].

Immature intermediate villi

As suggested by their name, immature intermediate villi are the immature continuations of stem villi that are in the process of development. They are characterised by a bulbous enlargement of their diameter to approximately twice the diameter of the foregoing stem [39]. Immature intermediate villi have a thick, uniform layer of trophoblast and are supplied by an arteriole and a venule [62, 68]. They are more prevalent in immature placentas as they are considered the growth centres of the villous trees during development of the placenta [68]. Although they are also found in mature placentas [68], they typically exist in small groups in the centre region of villous trees [66]. The peripheral branches of immature intermediate villi usually transform into mature intermediate villi while the older, more proximal ends undergo progressive stromal fibrosis to form stem villi [68]. The immature intermediate villi are likely the principal sites for material exchange during the first and second trimesters of human gestation [12].

Mature intermediate villi

Mature intermediate villi are the villous connections between the stem villi and the terminal villi. These elongated and gently curving intermediate villi have calibres ranging from 40 µm to 150 µm [12, 62, 66]. Mature intermediate villi have a thinner trophoblast bilayer compared to stem villi and contain evenly distributed fetal capillaries [62]. These structural properties suggest that mature intermediate villi may be involved in exchange of materials between the maternal and fetal compartments [12].

Terminal villi

Physiologically, terminal villi are the most important component of the villous tree. Terminal villi are located at the most peripheral region of the villous tree and typically arise as single or aggregated convolutes at intervals across the surface of mature intermediate villi [12]. Terminal villi have diameters of approximately 30 µm to 80 µm [12, 62, 63]. In normal mature placentas, terminal villi comprise nearly 40% of the volume of the placental villi and their total surface area accounts for approximately 50% of the total surface area of the villi [12].

Unlike stem villi, the trophoblast bilayer is thin and non-uniform over the villous surface [62, 66]. Terminal villi are predominantly occupied by fetal capillaries, which can take
up more than 35% of the villus volume [12]. This includes dilated capillary segments known as fetal sinusoids, which bulge against the thin trophoblast bilayer [12, 62], bringing the fetal bloodstream into close proximity to the maternal blood in the IVS. The thickness of villous membrane that separates the maternal and fetal bloodstream is as small as 0.5 \( \mu m \) to 2.0 \( \mu m \) [66]. These sites are known as vasculosyncytial membrane and their morphology enhances the efficiency and effectiveness of terminal villi for materno-feto exchange of materials.

### 2.2.3 Number and distribution of villous trees

There is a general consensus that the number of villous trees decreases as gestation advances [65, 69, 70]. Progressive regression of some villi has been observed from around 8 weeks of gestation, and the rate of regression slows down towards later stages of gestation [65].

Data on the number of villous trees in a mature placenta varies from one study to another. Due to inconsistent terminologies and difficulties in separating and correctly identifying a villous tree, some studies have provided a relatively higher estimate than other studies. For instance, Strauss [71] reported 1000 villi in a mature placenta and Crawford [70] counted 200 ‘fetal cotyledons’ (without providing a clear definition for fetal cotyledon), while other studies suggested a lower range of 40–70 villous trees in a mature placenta [61, 65, 66, 72]. It is not apparent how these studies derived these numbers, except for Boyd and Hamilton [65] who manually counted the villous trunks. However, as most studies estimated 40–70 villous trees [61, 65, 66, 72], this appears to be the currently accepted range of villous trees in a mature placenta.

### 2.3 The maternal uterine vasculature

Maternal blood flow to the placental bed and spaces surrounding the villous trees arises via the uterine circulation, which is supplied by the uterine artery and the ovarian artery. The uterus consists of three tissue layers: (1) the endometrium which lines the innermost uterine cavity, (2) the myometrium which is the thick muscular middle layer, and (3) the perimetrium which forms the outermost surface of the uterus [73]. Figure 2.3 illustrates the structure of the arterial system in the uterus. The uterine
artery and ovarian artery give rise to arcuate arteries after entering approximately a third of the myometrial thickness, which branch into radial arteries that traverse the myometrium towards the endometrium [73]. The radial arteries continue to branch into basal arteries and SAs as they advance through the myometrium. Basal arteries are mainly involved in supplying nutrients to the basal portion of the endometrium [65], while the highly coiled SAs penetrate towards the endometrial surface and open into the IVS surrounding the placental villi (Figure 2.2). The human placenta is categorised as a hemochorial placenta as the trophoblastic surface of the villi is directly bathed by maternal blood with the erosion of maternal blood vessels [12]. Maternal blood in the IVS is drained back to the main maternal circulatory system through the DVs, which coalesce and drain into the myometrial veins, uterine veins, ovarian veins, and finally into the internal iliac system [65].
Chapter 2. Background

Figure 2.4: The progression of SA remodelling through gestation. During pregnancy, SAs undergo remodelling whereby the tightly coiled structure (left) are invaded by placental trophoblasts which occlude the artery (middle) for the first 10–12 weeks of pregnancy. The invasive trophoblasts facilitate loss of elastic fibres and vascular smooth muscle cells, and transform the SA into wide bore conduits (right), rendering the artery independent of maternal vasoconstriction. Figure reproduced from James et al. [76].

2.3.1 Remodelling or changes in the uterine vessels during pregnancy

Spiral arteries

SAs which feed the IVS undergo remodelling as pregnancy progresses. One obvious difference between a pregnant and a non-pregnant uterus is that the terminal ends of SAs become open-ended and dilated during pregnancy. SA openings into the IVS are described as slit-like orifices with a funnel-shaped chamber [75] (right panel in Figure 2.4), where the diameter of SAs at the myometrial-endometrial junction and within the endometrium is approximately 200 µm [65], while the terminal end of an SA opening to the IVS at term ranges between 2–3 mm [65, 75].
2.3. The maternal uterine vasculature

The structural changes in SAs during pregnancy are brought about by a remodelling process known as ‘spiral artery remodelling’ [77]. During this process, invasive trophoblasts migrate in a retrograde manner through the lumen of the SAs by adhering to and replacing the endothelial cells of the SAs, whereby locally forming intraluminal trophoblast plugs [78, 79] (middle panel in Figure 2.4). It remains controversial whether blood flow into the IVS is completely obstructed by the trophoblast plugs in the SAs during the first 10 to 12 weeks of pregnancy [65, 80–83]. However, studies have confirmed that these plugs dissipate between 10 to 12 weeks and maternal blood flows freely into the IVS thereafter [84–86].

Decidual veins
Compared to the SAs, the DVs are generally less studied. These veins communicate freely with the IVS from early stages of pregnancy [65]. Trophoblast invasion or plugs are rarely observed in these veins [12, 65]. Apart from dilation of the venous lumens, these veins do not display the specific physiological changes like those observed in the SAs [77, 87].

Extra-decidual uterine vessels
Extra-decidual uterine vessels such as the radial, arcuate and uterine arteries, also undergo dilation throughout gestation especially if they are located beneath the placental implantation site [88, 89]. The dilation of these extra-decidual uterine vessels through pregnancy is significant, with the diameter of the uterine artery doubling by 6.5 weeks of gestation, and some arcuate arteries being twice as big as the uterine arteries at term [89]. As a result of these modifications, uterine blood flow increases from 750 mm$^3$/s (45 ml/min) in the non-pregnant state [90] to $8.33 \times 10^3 - 1.25 \times 10^4$ mm$^3$/s (500–750 ml/min) at term [55, 91–96].

2.3.2 Number and distribution of uteroplacental vessels

Spiral arteries
The exact number of SAs supplying a mature placenta is still unknown, due to different methodologies and difficulties in identifying and counting these vessel structures. The number of SAs thus varies between studies, with ranges of 40–50 [72, 97], 94–120 [66, 87, 98–100], and 180–320 [65, 101] estimated for a mature placenta.
Figure 2.5: A corrosion cast showing one to one relationship between an SA and villous tree. Maternal blood from the SA (red portion) is unobstructed and flows into an empty core of the villous tree (white portion) [102].

X-ray cineradiography, plastoid injection, radiography, and histomorphological studies have been employed to demonstrate a one to one relationship where a single SA supplies the central region of a villous tree [102–107] (Figure 2.5). However, Benirschke et al. [12] pointed out that this idealised arrangement is rarely observed throughout the placenta and tends to only occur in peripheral regions, where the average diameter of the villous tree is small. In the central region of the placenta, the villous trees are usually bigger and more developed. The trees in this region tend to overlap each other, which makes it difficult to differentiate the boundary of a villous tree to identify its orientation to the SA openings. Contrary to a central feeding SA, some investigators have also reported a random distribution of SA openings scattered over the basal plate [87, 88, 108, 109], while another group of researchers [110, 111] believed that the SAs open into the peripheral margin of each villous tree, which sometimes exist in groups of two or three near the base or in the lower third of the placental septa1 [87].

Decidual veins
The number of venous openings is uncertain as it is challenging to differentiate venous openings from arterial openings since venous openings may be dilated [65]. Some studies have suggested that there are less venous openings than arterial openings [65, 101, 111], for example, Klein [110] estimated 2–3 DVs for every 3–5 SAs. Most studies estimated that there are approximately 50–200 DVs in a term placenta [66, 99, 101], which is similar to the number of SAs, but again has a large range.

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1Placental septa are tissue projections that extend from the basal plate through the IVS towards the chorionic plate. The shape and size of septa vary considerably within a placenta and separate the placenta into lobes with each lobe containing one or more villous trees [112].
2.4 Maternal blood flow and its interaction with villous tree structure

Like the distribution of SAs, accounts of the distribution of venous openings on the basal plate are inconsistent between different studies. They have been reported to be evenly distributed by Arts [88] but Boyd and Hamilton [65] claimed a random distribution over the basal plate. The venous openings are thought to be located peripherally around each villous tree near the septa [66, 101, 107, 113]. However, in some studies [65, 99, 114, 115], the venous openings have shown a tendency to concentrate near the placental margin.

2.4 Maternal blood flow and its interaction with villous tree structure

With the loss of vascular smooth muscle cells [78] and elastic fibres from the vessel wall [74] during the remodelling process, the terminal end of the SAs lose vessel contractability, and blood flow from the maternal circulation into the IVS is no longer controlled by local vasomotor activity [65]. This guarantees the developing fetus a certain degree of nutrient and oxygen supply from the mother as the mother cannot restrict her nutrient and oxygen supply to the placenta, without reducing the supply to her own tissues [116].

While maternal blood flow to the uterus is distributed to both the placenta and the uterine tissue during pregnancy, there is no established technique that can reliably measure or estimate the distribution of flow between these two compartments [65]. Although the demand for blood supply in non-placental uterine tissue increases with its size during pregnancy [117], the mass of the placenta and its demand for blood supply increases even more rapidly [65]. Given that most of the maternal blood supplied to the placenta is channelled into the IVS during the second and third trimesters of pregnancy, it is reasonable to assume that the increase in uterine blood flow during pregnancy is indicative of the blood flow channelled into the IVS of the placenta [65].

As such, the maternal blood flow to a term placenta is estimated to be approximately $8.33 \times 10^3 - 1.25 \times 10^4$ mm$^3$/s (500–750 ml/min) [55, 91–96].

The rate of blood flow into the IVS is primarily controlled by (1) the flow impedance presented by the villous trees in the IVS, and (2) the pressure difference between the SAs and DVs [66]. In the IVS, villous trees are in close proximity with one another such that the tips of some trees fit into the notches of another tree leaving little space between villous trees [118]. Maternal blood in the IVS circulates through an interconnected
network of irregular and narrow cleft-like space within the bulk of the villous trees. These cleft-like spaces have been measured using scanning electron microscopy and are generally of capillary dimension [119] (approximately 16.4 µm to 32 µm [12]), and have been suggested to vary in size in response to the turgidity of fetal blood vessels which is controlled by the blood pressure in the fetal bloodstream [118].

An area of controversy revolves around the presence of a relatively villus-free cavity in the central region of villous trees (‘central cavity’). As our knowledge of the villous tree is derived from morphological studies performed on delivered placenta, only limited information is retrieved about the central cavity as the delivered placenta is devoid of maternal blood supply and lacks the physiological intraplacental blood pressure which maintains the villous trees in their in vivo state. Some researchers have argued that the central cavities observed in injection studies were due to insufficient filling of the vessels in the villous tree and they have demonstrated that the central cavity was absent in a specimen that was completely injected using a physiological pressure [60]. In another study [111], Grünwald attributed the small loose areas in the centre of villous trees in histological slides as artefacts and claimed that the SAs open into the IVS between villous trees.

However, X-ray cineradiography, plastoid injection and histomorphological studies [70, 102, 104, 120–122] revealed unobstructed flow of maternal blood into the empty core of the villous trees. As maternal blood pressure has been estimated to be approximately $9.33 \times 10^3 - 1.07 \times 10^4$ Pa (70–80 mmHg) at the level of the uterine artery, and approximately $1.33 \times 10^3$ Pa (10 mmHg) in the IVS [65], it is plausible that the high haemodynamic pressure of incoming maternal blood pushes aside the villi that lie in its path of flow, thereby creating a central cavity in the villous tree [72, 121]. The central cavity may be of haemodynamic significance [70] as it prevents maternal blood from ‘short-circuiting’ directly from the SAs to the DVs without penetrating the villous tree.

If the placenta is densely packed with interlacing villous trees without any empty cores, it would be difficult to explain how maternal blood could penetrate the thickness of the placenta using a propelling physiological blood pressure of only $1.07 \times 10^4$ Pa (80 mmHg) [121]. ‘Jets’ from the SAs into the IVS have also been visualised using colour Doppler ultrasonography [123] and a similar fountain-like spurt has also been reported in primate placentas [124]. This is consistent with the ‘physiological concept’ of placental circulation which describes maternal blood flow in the IVS [124]. In this theory, maternal blood ‘spurting’ into the IVS slows down as it heads towards the chorionic plate. Maternal
2.4. Maternal blood flow and its interaction with villous tree structure

blood disperses and seeps into the interconnected space between the densely packed villous branches, giving rise to a random combination of concurrent, countercurrent and cross-current flow of maternal blood between placental villi [125]. This flow orientation has been described as a multivillous flow system [126]. As maternal blood progresses through the IVS, it experiences impedance from the villous tissue and slows down further [124]. After traversing the villous tree, maternal blood reaches the peripheral region of the villous tree where the villi are more loosely packed and the intervillous clefts are relatively larger. These intervillous clefts drain into the relatively villus-free spaces in between trunks of the villous trees and close to the chorionic plate, which are known as the subchorial lakes. Back flow of the venous blood to the venous openings occur through the space between the villous trees as it offers the path of lowest blood flow resistance.

Wigglesworth [104] identified three different zones in the placental subunit. As illustrated in Figure 2.6, the central relatively villus-free area is an arterial zone (A), the dense meshwork of villi represents the capillary zone (C) and the peripheral region and region near the chorionic plate correspond to a venous zone (V). Simultaneous measurements at multiple locations in the IVS of a rhesus monkey placenta [127] also revealed the highest hydrostatic pressure and oxygen tension in the centre of a fetal cotyledon and lowest values in the subchorionic region, the periphery of the cotyledon and near the basal plate. Together with the confirmation of a central-to-peripheral gradient of oxygen distribution in other studies of primate placental subunits [128, 129], these studies provide evidence for the ‘physiological concept’ of placental circulation. Although accurate measurements of oxygen distribution are difficult to obtain directly from \textit{in vivo} human placenta, a central-to-peripheral oxygen distribution gradient in a placental subunit was inferred indirectly based on the activity of catalase (an antioxidant enzymes) in the placenta [130]. Recently, this oxygen distribution pattern has also been visualised in humans using blood oxygen level dependent imaging (BOLD)\(^2\) [131]. It is likely that the regions with low oxygen content (dark regions), which appeared as thin finger-like structures stretching from the fetal surface towards the maternal surface of the placenta, correspond to the peripheral regions of the placental subunit.

\(^2\)BOLD is an MRI technique that estimates regional changes in tissue oxygenation [131].
Chapter 2. Background

Figure 2.6: The three zones of maternal circulation described by Wigglesworth [104]. Oxygenated maternal blood enters into the IVS in the arterial zone (A). Oxygen exchange occurs as maternal blood flows past the villous tissue in the capillary zone (C), and oxygen-depleted maternal blood in the peripheral regions of the placental subunit and near the chorionic plate flows towards the DVs in the venous zone (V).

2.5 Oxygen and substrate exchange

Figure 2.7 shows the diffusion pathway taken by oxygen from the maternal bloodstream to the fetal bloodstream. Oxygen traverses a diffusion barrier consisting of five main compartments in series: (1) maternal erythrocytes (me), (2) maternal blood plasma (mp), (3) villous membrane (vm) which includes the trophoblast bilayer, stroma and fetal capillary endothelium, (4) fetal blood plasma (fp), and (5) fetal erythrocytes (fe). Each of these compartments provides resistance to oxygen diffusion, and the total diffusing capacity of oxygen in the placenta is the summation of the reciprocal of conductance in each compartment. Studies have revealed that exchange efficiency is most dependent on the diffusing capacity of the villous membrane among the five compartments [132, 133]. As the diffusing capacity of the villous membrane is determined by the mean harmonic thickness of the villous membrane, villous surface area, and fetal capillary surface area, placental structure plays an important role in oxygen and substrate exchange in the placenta. Other than the diffusion barrier thickness, the amount of oxygen that the fetus can extract from the maternal circulation is also affected by factors such as uterine and umbilical arterial oxygen partial pressures, maternal and fetal blood flow rates, and vascular arrangement of fetal blood vessels to maternal blood flow [134].
2.6 Placental pathology and its relationship to placental structure

Since the placenta is the only interface that supplies oxygen to the fetus during pregnancy, pregnancy complications have often been linked to the malfunction of placenta [135, 136]. In this section, we explore the pathology of the placenta, with particular focus on IUGR and pre-eclampsia.

2.6.1 Intrauterine growth restriction

IUGR is the failure of the fetus to reach its optimal growth potential and is a leading cause of perinatal morbidity and mortality [137, 138]. Fetuses with IUGR are at a higher risk of stillbirth, neonatal morbidity, and mortality [139–141]. Clinically, IUGR can be categorised as symmetric or asymmetric growth restriction.

Fetuses with symmetric IUGR generally display overall symmetrical growth impairment [139] which usually arises from prolonged intrinsic insult from chromosomal abnormalities, congenital malformations, drugs, or infections [139]. Compared to normal placentas, villi
in IUGR cases are elongated, sparse, and poorly developed [18] (top panel in Figure 2.8). It is likely that these structural abnormalities increase fetoplacental vascular impedance at the fetal capillary level, which is consistent with the reduced or absent end-diastolic flow velocity that is frequently observed in the umbilical arteries of fetuses diagnosed with symmetric IUGR [135]. This leads to impaired oxygen uptake by the fetus, resulting in a hyperoxic IVS; in these cases, it has been found that the oxygen content of maternal blood exiting the IVS is close to the content in SAs [142]. The hyperoxic environment may result in the loss of hypoxic drive for trophoblastic proliferation and angiogenesis, and is likely to further perpetuate impoverished villous development and suboptimal oxygen uptake [135]. It remains unclear which is the initiating factor of this vicious cycle [135].

Fetuses with asymmetric IUGR have relatively normal head circumference but their abdominal circumference is usually smaller than expected [139]. Unlike symmetric IUGR, asymmetric IUGR presents later in pregnancy and commonly arises from disorders such as pre-eclampsia [139]. There is evidence of reduced uterine blood flow to the placenta in asymmetric IUGR as demonstrated by radio-isotope studies [144], colour Doppler imaging [145] and MRI [146]. It is speculated that reduced uterine blood flow to the placenta provides a hypoxic trigger for increasing development of the villous tree [135] (bottom panel in Figure 2.8), which may explain why asymmetric IUGR is associated with normal umbilical artery Doppler waveform [147], accelerated maturation of the placenta [148], and an increase in capillary volume fraction compared to normal placentas [149].

2.6.2 Pre-eclampsia

Pre-eclampsia affects about 5–8% of pregnant women worldwide and presents with symptoms such as hypertension and proteinuria [136]. Early-onset pre-eclampsia (<34 weeks of gestation) is often characterised by alterations in uterine Doppler artery profiles [150], which suggests inadequate uterine SA remodelling. In up to one-third of the cases, early-onset pre-eclampsia is accompanied by IUGR [138]. The volume and surface area of terminal villi have been reported to be compromised in these early-onset cases [16], but these abnormalities have been attributed to IUGR [66] as their morphologies resemble placentas from IUGR cases without pre-eclampsia [151]. In some cases, women only present symptoms during late pregnancy (>34 weeks of gestation) [136] and these women usually have a larger placental mass or surface due to conditions such as diabetes,
2.6. Placental pathology and its relationship to placental structure

Figure 2.8: Morphology of villus affected by pregnancy complications. Terminal villi are elongated in IUGR cases with absent end-diastolic flow in umbilical artery (AEDFV) while increased branching is observed in IUGR cases and pre-eclampsia cases with normal end-diastolic flow in umbilical artery (EDFV). Oblique lines across villi refer to the location where the cross sections are taken. Modified from Kingdom and Kaufmann [143].

multiple pregnancies, anemia and high altitude [150]. Unlike early-onset cases, the Doppler waveform of uterine arteries [150], placental weight, volume, morphology and umbilical artery flow Doppler profile are generally normal in late-onset pre-eclampsia, and the fetuses are usually not growth restricted [66, 136, 150].
2.7 Existing computational models and knowledge gap

To better understand placental physiology in normal and pathological pregnancies, numerous computational models have been developed to investigate the structure and/or function of the placenta. Most studies have placed particular emphasis on modelling the transfer of gas between the maternal and fetal bloodstreams in placentas from different species [48–52, 152, 153]. For example, the sheep placenta was modelled as a black box [49], and the oxygen transfer efficiency was estimated from measurements of oxygen entering and leaving the placenta. However, the contributions of various placental structures to its function cannot be elucidated.

To examine the structure-function relationship of the placenta, capillary-scale placental models idealised the fetal and maternal bloodstreams into two compartments separated by an exchange barrier made up by a placental membrane layer [50, 52, 152, 154–157]. Systems of differential equations were applied to model diffusive mass transfer across the placental barrier based on Fick’s law. Although most models assumed a simple case of concurrent flow [50, 52], different maternal blood flow direction in relation to the fetal blood flow can be implemented to investigate how different orientations influence exchange kinetics. For instance, Faber [48] used an advection-mass transfer model and identified that the counter-current flow pattern, typically found in guinea pig and rabbit placentas, is the most efficient exchanger among counter-current, concurrent, cross-current (maternal capillaries are perpendicular to fetal capillaries), pool flow (a uniform solute concentration along the length of maternal or fetal capillaries), and double pool flow patterns (uniform solute concentrations in both maternal and fetal capillaries) (Figure 2.9). However, as the multivillous flow pattern in the human placenta is not appropriately captured by any of these five patterns, findings from this model have limited implications for understanding exchange efficiency of the human placenta.

To capture the flow orientation in the human placenta, Guilbeau and Reneau [155] modelled the idealised hair-pin structure of a fetal vessel in a terminal villus by connecting a concurrent flow exchange unit with a counter-current flow exchange unit to simulate concurrent maternal blood flow with the fetal circulation on one side and a counter-current flow on the opposite side of the villus. However, in this model and many other dual compartment models [52, 152, 155], the diffusion path of oxygen is often oversimplified,
2.7. Existing computational models and knowledge gap

Figure 2.9: Patterns proposed by Faber [48] for placental flow in different animal species. Among the five flow patterns, the countercurrent pattern is most efficient, but none of the patterns adequately describes the multivillous flow pattern observed in the human placenta.

with limited consideration for precise morphometric data, the effects of haemoglobin levels, and the balance of maternal to fetal blood flow on exchange kinetics.

Laga et al. [132] and Mayhew et al. [133] attempted to better describe the diffusion path of oxygen by expressing the diffusing capacity of oxygen in terms of the geometrical characteristics of the villi and the IVS measured from the human placenta. Unlike these models which used averaged morphometric measurements [132, 133], Gill et al. [158] developed a model which calculates oxygen diffusion using actual geometries of terminal villi segmented from digital photomicrographs of terminal villi. Although this model allows quantitative assessment of the contribution of various biological distances in the villi to oxygen transport, oxygen content of the maternal bloodstream was assumed constant everywhere, and the significant spatial variations of maternal oxygen concentration in the IVS were totally ignored. These morphometric diffusing capacity models also disregarded the flow dynamics of fetal blood circulation by assuming perfect-sink conditions at the villi boundaries.

Most feto-placental blood flow models are based on the chorionic vessels which have been studied extensively in morphometric studies due to their accessibility [12, 159, 160]. Despite the wealth of information on the complex network of chorionic vessels, simplification of the geometry remains inevitable for computational tractability. For
example, Gordon et al. [161] only examined flow in a small subset of approximately five chorionic vessels using computational fluid dynamics techniques, while Franke et al. [162] neglected arterial curvature, and assumed axisymmetry in flow to solve for 1D blood flow in the entire chorionic tree. In these models, feto-placental vessels beyond the chorionic vessels (i.e. villous vessels) were not explicitly represented and were replaced with pressure or flow boundary conditions at the inlet to each villous tree.

With the advent of imaging techniques such as confocal microscopy and micro-computed tomography (micro-CT) imaging [163, 164], the structure of the villous vessels have been studied more extensively. Using data acquired from micro-CT imaging of mouse placental arteries, Yang et al. [164] attempted to include several generations of villous vessels in their model. However, their model only accounts for vessels which can be visualised using micro-CT, while high-resistance vessels that were undetected due to the resolution limit of micro-CT were neglected in this model. It is also a challenge to determine a physiological representation for the boundary conditions assigned at the terminal vessels. Very often, the size of blood vessels at which boundary conditions must be applied is too small for visualisation by the current available imaging techniques and metrics such as flow and pressure at such a level are also difficult to measure.

A way to circumvent these limitations is to consider the feto-placental vasculature from the umbilical arteries to the umbilical vein. Boundary conditions can be readily defined with the flow and pressure in the umbilical arteries (inlet) and vein (outlet) using flow velocity waveforms measured at the umbilical vessels. However, given the huge number of vessels in the entire feto-placental vasculature, simplification of the geometry and/or flow in the model is necessary for computational tractability. In a study by Thompson and Trudinger [165], the branching structure of the fetal circulation was reduced to two levels of branching arteries with a large number of parallel branches at each level, where the first level represents primary and secondary stem villi, while the second level corresponds to the tertiary villi. In other studies [166, 167], the asymmetric structure of the feto-placental vasculature was simplified to a dichotomous symmetric tree with 14–15 branching generations. In these models, properties such as the number of branching generations, vessel calibre as well as the number of terminal branches were varied to investigate their influence on fetal blood flow resistance and flow rate. Although these models predicted dependency of fetal blood flow on the structure of the vessel tree,
key structure-function relationships may be compromised by the extensive geometrical simplification of the feto-placental circulation.

A recent multiscale model has attempted to generate an anatomically accurate feto-placental vasculature across the different spatial scales by incorporating the key structural descriptions of an ‘average’ placenta gathered from the literature [168]. The model included six to eight generations of chorionic vessels branching from the point of umbilical cord insertion to supply 60–100 villous trees. Each villous tree was in turn represented by another 15 generations of branching and fitted to match the literature descriptions as closely as possible. The model also included a fetal venous system that follows the arterial system. Using Poiseuille flow to model blood flow through this complete feto-placental circuit, the size and number of vascular branches have been shown to affect feto-placental resistance, flow heterogeneity, and placental efficiency. However, as no attempt was made to include maternal blood flow, the model cannot be applied to study the influence of maternal blood flow on fetal oxygen uptake.

To capture the characteristics of the flow field within and surrounding a villous tree, the human placenta has been conceptualised as a porous medium [53, 54]. The dense meshwork of villous trees in the placenta has been represented by the matrix or skeleton of the porous medium, while the maternal blood flow that infiltrates the spaces within the villous tree is modelled with Darcy’s law [53, 54]. These studies were interested in simulating the flow dynamics which occur as a result of the ‘obstructions’ by the villous trees. As shown in Figure 2.10a, Erian et al. [53] modelled the villous tree as a square domain with: (1) a constant permeability, (2) two zones of different permeabilities with a higher permeability in the central region which represents the relatively villus-free central cavity, and (3) a medium in which the local permeability increased with local flow speed to mimic the effects of villous tree deforming and spreading out in response to maternal blood spurts. Results from this model demonstrated ‘short-circuiting’ of maternal blood flow from the SA directly to the DVs without penetration into the depth of the placenta but this has been attributed to the lack of consideration of flow inertia and pulsatility of maternal blood flow in the model.

Chernyavsky et al. [55] proposed an improved model which assumed that a mature villous tree takes the form of a hemisphere with a central cavity, which represents the villus-free core region of the villous tree (Figure 2.10b). Chernyavsky et al. [55] continued
Figure 2.10: Schematic representation of existing placentone models. (a) Erian et al. [53] proposed a 2D placentone represented by a square domain filled with porous medium, which is supplied by a central SA and drained by two DVs. As the central cavity (region bound by two dashed lines) is relatively villus free, it was assigned a higher permeability than the region surrounding the central cavity. (b) Schematic of the 3D placentone model developed by Chernyavsky et al. [55]. The villous tree was represented as a porous medium in a hemispherical domain with an SA feeding directly into the central cavity and DVs positioned at the peripheral of the placentone. (c) Maternal blood flow from the central SA to the DVs was modelled as stream tubes in the model proposed by Serov et al. [56].

to use Darcy’s law to describe maternal flow, and derived the flow field and pressure field of maternal flow through the field of the IVS. Without accounting for flow inertia in the model, the model demonstrated that oxygen delivery to the placenta improves with increased flow penetration into the placentone when the DVs are located near the periphery of the placentone. Although Chernyavsky et al. [55] took a step towards representing the gross geometry of a mature villous tree in 3D, this model remains a lumped parameter model. Such homogenisation is not truly reflective of the complex and inhomogeneous branching villous tree structures in human placentas and the model is unlikely to capture the influence of fine-scale geometric features of the villous tree on IVS flow dynamics and oxygen exchange efficiency. In another model, Chernyavsky et al. [169] simulated flow and diffusion around distributed point sinks and performed statistical analysis of 2D placental sections to estimate the potential errors in the homogenisation of IVS tissue to uniform or slowly varying area fractions. They showed that the accuracy of homogenisation techniques depend on flow characteristics (Péclet and Damköhler numbers), as well as the relative size of intervillous distances and typical pathlengths from SAs to DVs. Recently, a two-dimensional model of flow in the IVS developed by Lecarpentier et al. [170] represented the villous tree using a corrected cross-sectional image obtained between the chorionic plate and the basal plate of a normal placenta in
the third trimester, and modelled maternal blood flow around ‘rigid’ villous tissue in the IVS using Navier-Stokes equation. While this approach includes significant geometric details, it is computationally expensive for large scale simulations of blood flow and/or nutrient transport in the whole placenta.

Serov et al. [56, 57] adopted a different approach by introducing a stream-tube model of oxygen exchange in a placental subunit. The model assumed that maternal blood flow streamlines follow villous branches that are uniform along their length [56] (Figure 2.10c). These geometric simplifications allow analytical (therefore rapid) solution of models to predict function, and can be parameterised by assessment of histological slides showing the size and distribution of villous tissue. These models therefore provide estimates of the efficiency of the placenta but do not account for the branching architecture of the villous tree, which may significantly influence placental function [171].

To bridge the gap between detailed geometric models [168, 170] and highly smoothed homogeneous models [55–57], we developed a computational model of blood flow and material transport in a human placental subunit to include the well-defined branching structure of the villous tree and a smoothed representation of the terminal villi. In the next chapter (Chapter 3), we will describe our novel maternal blood flow model, with specific details on how we define a non-uniform porosity distribution using the key structural features of a villous tree, and how we simulate maternal blood flow in the resulting porosity field to ultimately investigate the effects of villous tree geometry on maternal blood flow.
Chapter 3

Predicting maternal blood flow in a placental subunit

3.1 Motivation

Here, we present a composite model of maternal blood flow in the IVS, which aims to bridge the gap between detailed geometric models [168, 170] and highly smoothed homogeneous models [55–57], providing a picture of how the architecture of the placental villi influences maternal blood flow in the placenta. Our aim is to also use the model in later chapters to interpret MRI images showing placental blood flow and to understand how villous tree geometry influences distribution of oxygen in the IVS and fetal oxygen uptake rate. Work discussed in this chapter has been published [172].

3.2 Methods

A schematic of model components and key outputs is given in Figure 3.1, and each component is described below. This modelling framework is gestation stage-independent and can be parameterised to different stages of gestation. We solved these models using a custom written Matlab® code (The Mathworks Inc., version 2012b).
1. Set up domain

2. Generate villous tree structure

3. Define sampling grid

4. Fill sampling grid elements with terminal tissue blocks

5. Fit \( \kappa \) field using sampling grid elements as data points

6. Predict maternal blood velocity field in IVS
   a. Pressure distribution
   b. Flow streamlines

**Figure 3.1:** Components of the maternal blood flow model. The steps involved setting up a domain with a villous tree structure and superimposing a sampling grid over the villous tree to determine a conductivity field that varies with the structural properties of the villous branches and terminal tissue blocks. Maternal blood flow in the resultant conductivity field was solved to determine the flow streamlines and pressure distribution in the IVS.
3.2. Methods

3.2.1 Model geometry

Construction of a domain

A placental subunit was represented either by a 2D or 3D domain, with \( n_{SA} \) SAs and \( n_{DV} \) DVs on the basal plate of the domain, and \( n_{villous} \) villous trees positioned on the opposing chorionic plate.

Representation of villous tree

The villous structures to the level of the intermediate villi were considered as distinct branches in this model, but beyond that level the terminal villous structures become too numerous to be explicitly included in a model, with a branching structure that is not well described \[168\]. Like in previous models of villous vascular branching \[168\], these were lumped together as a ‘terminal unit’.

Branching component in 2D models

A rule based approach was implemented to generate tree structures in 2D models (Figure 3.1, step 2). A starting branch of length \( l_s \) and diameter \( d_s \) was positioned so that the generated tree stemmed from the chorionic plate of the domain with an axis of symmetry aligned with the opening of the SA. It was assumed that new branches emerge only from existing branch tips. Before branching can occur, the following conditions had to be satisfied:

1) There must be sufficient space locally for a new branch to form. Specifically, a new branch cannot grow outside the boundary of the domain or into space already occupied by another branch.

2) The branching generation of the new branch is below the predefined total number of branching generations (\( n_b \)).

If both conditions are satisfied, the new branch will grow according to the dimensions specified by the daughter to parent branch length ratio \( (l_d/l_p) \), diameter ratio \( (d_d/d_p) \), and branch angle \( (\theta_b) \).

With this algorithm, large voids between the domain boundaries and the stem would occur if the villous tree was defined with a uniform branch angle across all generations. This was avoided by manipulating the branch angle for the first 4 generations so that
Table 3.1: Branch angles for villous tree in 2D models.

<table>
<thead>
<tr>
<th>Branching generation</th>
<th>Angle between parent and left branch</th>
<th>Angle between parent and right branch</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>$35^\circ$</td>
<td>$35^\circ$</td>
</tr>
<tr>
<td>2</td>
<td>$70^\circ$</td>
<td>$10^\circ$</td>
</tr>
<tr>
<td>3</td>
<td>$15^\circ$</td>
<td>$45^\circ$</td>
</tr>
<tr>
<td>4</td>
<td>$7^\circ$</td>
<td>$45^\circ$</td>
</tr>
<tr>
<td>5 &amp; above</td>
<td>$18^\circ$</td>
<td>$18^\circ$</td>
</tr>
</tbody>
</table>

The domain space was completely filled. The branch angles for the first 4 generations of the villous tree are shown in Table 3.1.

At each bifurcation, the branch nearer to the middle of the domain was given precedence over the other branch in order to create a tree with branches that grow towards the SA. The algorithm was implemented to create a tree occupying half of the domain, and reflected to obtain full tree symmetry. These parameters and conditions define a bifurcating tree as shown in Step 2 of Figure 3.1, where the most distal villi represent the intermediate villi.

Branching component in 3D models

For 3D models, a volume-constrained branching algorithm was used to construct the branching component of the villous tree. The algorithm was previously used to create morphometrically realistic pulmonary airway and vascular trees in the lungs and the placenta [168, 173]. The algorithm controls the number of branching generations ($n_b$) and branching asymmetry of the tree using the following parameters: the number of seed points ($n_{seed}$), stem length ($l_s$), stem diameter ($d_s$), the minimum length of an individual branch ($l_{min}$), the maximum branch angle of a daughter branch from its parent branch’s axis ($\theta_{max}$), the fractional distance that each branch grows to the centre of mass ($f_{dist}$), and the daughter to parent branch diameter ratio ($d_d/d_p$), as detailed in the following steps as shown in Figure 3.2:

1) Seed data points were distributed evenly over the volume of the 3D domain.

2) An initial branch of length $l_s$ and diameter $d_s$ was positioned on the chorionic plate with its stem aligned directly under the opening of the SA.

3) The seed data points were split into two groups with a plane defined by a line between the growth tip of the stem and the centre of mass of the seed data points, and a vector in the direction of the parent branch.
3.2. Methods

![Figure 3.2](image)

**Figure 3.2:** Generation of tree structure in 3D models. (a) The centre of mass (red dot) of the evenly distributed seed points (grey dots) was identified. (b) The seed points were divided into two sets by a plane defined by the centre of mass and the growth tip of the parent stem (grey cylinder). (c) The centre of mass of the subsets was calculated and daughter branches grew from the parent’s growth tip towards the new centre of mass by a predefined distance.

4) The centre of mass of the two new subregions was calculated. New daughter branches were grown a distance from the growth tip of the parent branch to the two centres of mass as defined by $f_{\text{dist}}$. The diameter of the daughter branch was calculated from $d_d/d_p$ using the diameter of the parent branch.

5) The length of the new daughter branches and the angle between the daughter and parent branches were checked to ensure that they were within the prescribed limits of $l_{\text{min}}$ and $\theta_{\text{max}}$. The branch angle was either reduced to satisfy the predefined limits or the branch was considered as a terminal branch, which led to the termination of the growth algorithm for this group of seed points.

6) Steps 3–5 were repeated continuously with progressively smaller sets of seed points. The algorithm was terminated when a minimum branch length was attained or when there was only one seed point left in each group.

The input parameters were altered successively to obtain a branching tree with properties that closely match $n_b$, $l_s$, $l_d$, $\theta_b$, and Strahler branching ratio\(^1\) as reported in the literature. As a variety of $n_{\text{seed}}$ and $d_d/d_p$ could give rise to a tree that satisfies these requirements,

\(^1\)Strahler branching ratio is an index of asymmetry and is calculated by counting the number of branches in each Stahler order ($b_N$) and determining the gradient of the straight line fitted to $\log(b_N)$ against order. A description of how to assign Stahler order to tree branches is provided by Strahler [174].
an additional fitting step was implemented to obtain a tree structure that closely matches
the villous volume estimated from stereological studies.

Beyond the intermediate villi
The convoluted terminal villi beyond the intermediate villi were modelled as homogenised
‘terminal tissue blocks’ (TBs), with villous tissue density as measured from stereological
studies that investigated the morphological properties of terminal villous tissue. To
do this, we superimposed a sampling grid over the tree structure (Figure 3.1, step 3),
and each element of the sampling grid that contains intermediate villi was assigned as
a TB with a villous tissue area fraction ($\phi_{TV}$), and an average villous diameter ($d_{TV}$)
(Figure 3.1, step 4). To provide a size that is representative of the space occupied by
terminal villous tissue arising from each intermediate villus, the size of elements in the
sampling grid (and so the size of each TB) was estimated using

$$\frac{n_{TV} \times l_{TV} \times d_{TV}}{\phi_{TV}},$$  \hspace{1cm} (3.1)$$

where $n_{TV}$ is the number of terminal convolutes branching from a mature intermediate
villus, and $l_{TV}$ is the cumulative length of a terminal convolute branching from an
intermediate villus. As terminal villi are typically observed only in the third trimester
[12], this component is not necessary for modelling placentas prior to that stage.

3.2.2 Maternal blood flow in the IVS

_Governing equations_
Explicitly modelling the fluid dynamics of maternal blood in the IVS between the
numerous generations of villi would be computationally impractical even in a placental
subunit. Therefore, we assumed that the IVS can be represented as a porous medium
[53–55]. However, we defined hydraulic conductivity ($\kappa = \kappa(x, y, z)$) as a spatially varying
quantity calculated from the generated tree and tissue structure (Figure 3.1, step 5). Maternal blood flow in the IVS was therefore modelled using Darcy’s law:

$$\nabla \cdot \mathbf{V} = 0,$$  \hspace{1cm} (3.2a)$$
$$\mathbf{V} = -\frac{\kappa}{\mu} \nabla P,$$  \hspace{1cm} (3.2b)$$
where $V$ is the blood velocity in the IVS, $\mu$ is the viscosity of maternal blood, and $\nabla P$ is the local maternal blood pressure gradient.

**Finite element mesh and solution procedure**

The conventional way to solve the system of equations 3.2 is to substitute Equation 3.2b in Equation 3.2a and determine the distribution of blood pressure in the domain using the standard Galerkin finite element method. The velocity field can then be calculated from Equation 3.2b using the finite difference method. However, this method usually results in fluid fluxes with discontinuous normal components across adjacent element interfaces [175]. Especially when applied to a field with inhomogeneous hydraulic conductivity, these discontinuities in normal components give rise to inaccurate fluid velocities [176] which would affect the computation of material transport in the domain [177].

To overcome the shortcomings of solving Darcy’s equations with the Galerkin finite element method, we used the mixed finite element method (MFEM) to simultaneously solve Equations 3.2a and 3.2b for both pressure and velocity fields by representing each of the unknowns with its own space of basis functions as detailed in Appendix A [175, 178, 179] (Figure 3.1, step 6).

As shown in Figure 3.1, the model output includes a pressure distribution and a velocity field in the IVS. The generated pressure is piecewise constant over each finite element mesh element while the velocity components are represented by the normal fluxes at each mesh element edge, which are continuous between adjacent elements.

**Boundary conditions**

When the model geometry was axisymmetrical, pressure boundary conditions were prescribed at the inlet SA ($P = P_{in}$) and outlet DVs ($P = P_{out}$). $P_{in}$ and $P_{out}$ were fitted to obtain a flow velocity at the inlet element edge that matches literature estimates of volumetric flow from a single SA ($Q_{in}$). The two pressures were then held constant between simulations. When the model geometry was asymmetric, pressure boundary conditions were inapplicable and a flow velocity matching $Q_{in}$ was applied as a boundary condition at the inlet element edge. As the basal and chorionic plates act as barriers to flow, the normal component of velocity was set to zero at these boundaries except at the inlet and outlets. At the boundaries between placental subunits, we applied a symmetry condition ($U(0,y) = U(\epsilon, y)$, where $\epsilon$ is the width of the placental subunit).
Derivation of a hydraulic conductivity field

The hydraulic conductivity coefficient ($\kappa$) in Equation 3.2b, was expressed using the Kozeny-Carman formula \[180\]:

\[
\kappa = \frac{d_{\text{villi}}^2 (1 - \phi)^3}{180 \phi^2},
\]

(3.3)

where $d_{\text{villi}}$ is the average diameter of villi in the IVS and $\phi$ is the tissue density of villous tissue. Each element in the sampling grid was assigned a unique $\kappa$ value, which represents the hydraulic conductivity of the tissue branches in that element or TB. As the branches within a sampling element are considered randomly oriented, we assumed that $\kappa$ is isotropic in all elements. The contribution to $\kappa$ of the TBs was defined by $\phi_{TV}$ and $d_{TV}$. The contribution of discrete villous branches in each element was defined using a weighted average of the diameter of villi in the element and the area occupied by branches in 2D models or the volume occupied by branches in 3D models. The maximum value of $\kappa$ in each element was prescribed a value of $\kappa_{\text{empty}}$. A smooth spatially varying field for $\kappa$ (Figure 3.1, step 5) was obtained for each element of the maternal flow mesh by sampling $\kappa$ at each element of the sampling grid (Figure 3.1, step 3) and averaging $\kappa$ over all elements surrounding each node of the sampling grid. Smoothing was then implemented via linear interpolation to determine $\kappa$ at the midpoint of each maternal flow mesh element based on the generated nodal data, and the resultant value was used as a $\kappa$ value for the maternal flow mesh element.

### 3.2.3 Model parameterisation

In this chapter, we parameterised the model in 2D for a term placenta, with nominal model parameters and physiological ranges from the literature listed in Table 3.2. The domain can take any shape but here we followed previous studies \[53\] and assumed a 2D rectangular domain. This is valid if one assumes that the placenta is of uniform thickness in the region of a villous tree. As shown in Figure 3.3, the placental subunit was represented by a domain with a thickness $\tau$, which spans the distance between the basal plate and the chorionic plate of the placenta, and a width $\epsilon$. While $\tau$ and $\epsilon$ were chosen following Chernyavsky et al. \[55\], estimates for placental dimensions vary in the literature. *Ex vivo* and *in vivo* estimates of placental volume are quite consistent at term \[65, 181\], although due to the loss of placental turgidity after delivery, placental
3.2. Methods

Figure 3.3: 2D model representation of a term placental subunit. The subunit was represented by a rectangular domain of thickness $\tau$ and width $\epsilon$. Each subunit contains a representative villous tree generated from realistic morphometric parameters fed by a central SA and drained by two DVs. The most distal branches of the villous tree shown are termed intermediate villi, and these villi were assumed to supply ‘terminal tissue blocks’.

thickness estimates vary between studies [65, 182]. For a given placental volume, the value of $\epsilon$ is expected to vary based on the assumption of a value for $\tau$.

Due to the lack of quantitative assessment of the distribution of the maternal vasculature with respect to the villous trees, we followed previous studies [53, 55–57] and assumed that the placental subunit is fed by one central SA and drained by two DVs. The distance between the SA and DV ($x_v$) was chosen based on a previous model [55], where an optimum delivery of nutrients was predicted when DVs are located near the periphery of the placental subunit. The SA and DVs were assumed to have a fixed diameter $d$.

The branching component of the villous tree has been described in several studies [12, 39–43, 63, 183]. For a term placenta, the villous branches at the interface between the chorionic plate and the villous trees have a length of approximately 2 mm and a diameter of up to 1.7 mm [63, 183], which branch through up to 15 generations to numerous intermediate villi whose diameter is approximately 60 $\mu$m [63], and finally the main contributors to gas exchange - terminal villi - form convoluted structures in which fetal capillaries come in close contact with maternal blood [12, 184]. Using these nominal values for a term placenta (Table 3.2), the model generated a resulting 2D villous tree structure comprised of 15 branching generations with 202 terminal units.
Due to differences in placental samples and/or experimental methods, estimation of the terminal villus density ($\phi_{TV}$) varies between studies [14, 16]. The tissue density estimated by Egbor et al. [16] was used as the nominal value as the study also provided morphometric data for a variety of pathological conditions, which can be applied to understand pathology. The diameter of terminal villi ($d_{TV}$) is generally within the range of 0.03–0.06 mm [16, 63] and hence, a mid-range value of 0.05 mm was assigned as the nominal value. At term, the cumulative length of a terminal convolute branching from an intermediate villus has been estimated to be $l_{TV} = 3$ mm, and there are approximately $n_{TV} = 8–10$ terminal convolutes branching from a mature intermediate villus [63]. Given that $\phi_{TV} = 0.4$, we implemented a sampling grid size of 2 mm × 2 mm at term.

Hydraulic conductivity is infinity in an empty sampling grid element but such infinity data points will result in sharp fronts during smoothing which are physically impossible, and difficult to deal with computationally. To overcome this, the maximum $\kappa$ calculated by superimposing the sampling grid over a term villous tree was scaled up by 10 times and set as the nominal value for $\kappa_{\text{empty}}$ for all simulations.

We assumed that there are 100 SAs as estimated by Lyall [100]. Using an estimated total maternal blood flow of $8.33 \times 10^3–1.25 \times 10^4$ mm$^3$/s (500–750 ml/min) in a term placenta [55, 92–95], $P_{in}$ and $P_{out}$ were fitted to obtain a value of 83.3 mm$^3$/s (5 ml/min) for $Q_{in}$. For the 2D model, the sampling grid is refined once to obtain a finite element mesh with an element size of 1 mm × 1 mm (the maternal flow mesh). The maternal flow mesh was further triangulated to obtain a mixed finite element mesh and the lowest Raviart-Thomas MFEM was implemented to solve Equation 3.2 (Figure 3.1, step 6).

### 3.3 Results

#### 3.3.1 Baseline case and consistency with previous studies

*Flow streamlines from baseline case*

Using the nominal parameter values, the pressure difference between the inlet and outlets was set to 2506 Pa (18.8 mmHg) to generate an average volumetric flow of 83.3 mm$^3$/s (5 ml/min) at the inlet. Figure 3.4 illustrates the predicted blood flow streamlines in the IVS. In the baseline case, blood flow streamlines are consistent with the previous
Table 3.2: Model geometry and maternal blood flow model parameters for a 2D term placental subunit. The table also includes chosen nominal value and literature range for each parameter.

<table>
<thead>
<tr>
<th>Symbol</th>
<th>Description</th>
<th>Nominal value</th>
<th>Literature range</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>( \tau )</td>
<td>Domain thickness</td>
<td>20 mm</td>
<td>20–45 mm</td>
<td>[12, 55, 65, 182]</td>
</tr>
<tr>
<td>( \epsilon )</td>
<td>Domain width</td>
<td>40 mm</td>
<td>10–40 mm</td>
<td>[12, 55]</td>
</tr>
<tr>
<td>( n_{SA} )</td>
<td>Number of SAs</td>
<td>1</td>
<td>1</td>
<td>[53, 55–57]</td>
</tr>
<tr>
<td>( n_{DV} )</td>
<td>Number of DVs</td>
<td>2</td>
<td>2</td>
<td>[53, 55–57]</td>
</tr>
<tr>
<td>( n_{villous} )</td>
<td>Number of villous trees</td>
<td>1</td>
<td>1</td>
<td>–</td>
</tr>
<tr>
<td>( x_v )</td>
<td>Distance between SA and DV</td>
<td>18 mm</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>( d )</td>
<td>Diameter of SA and DVs</td>
<td>2 mm</td>
<td>2–3 mm</td>
<td>[104, 185]</td>
</tr>
<tr>
<td>( n_b )</td>
<td>Number of branching generations</td>
<td>15</td>
<td>up to 15</td>
<td>[63, 183]</td>
</tr>
<tr>
<td>( l_s )</td>
<td>Stem length</td>
<td>2 mm</td>
<td>2 mm</td>
<td>[63]</td>
</tr>
<tr>
<td>( d_s )</td>
<td>Stem diameter</td>
<td>1.7 mm</td>
<td>1.7 mm</td>
<td>[63]</td>
</tr>
<tr>
<td>( l_d/l_p )</td>
<td>Daughter/parent branch length ratio</td>
<td>0.975</td>
<td>1</td>
<td>[63]</td>
</tr>
<tr>
<td>( d_d/d_p )</td>
<td>Daughter/parent branch diameter ratio</td>
<td>0.80</td>
<td>~0.80</td>
<td>[63]</td>
</tr>
<tr>
<td>( \theta_b )</td>
<td>Branch angle</td>
<td>18°</td>
<td>40°–70° (terminal branches)</td>
<td>[186]</td>
</tr>
<tr>
<td>( \phi_{TV} )</td>
<td>Terminal villi area fraction</td>
<td>0.40</td>
<td>0.28–0.40</td>
<td>[14, 16]</td>
</tr>
<tr>
<td>( d_{TV} )</td>
<td>Terminal villi diameter</td>
<td>0.05 mm</td>
<td>0.03–0.06 mm</td>
<td>[16, 63]</td>
</tr>
</tbody>
</table>

Maternal blood flow model

\[
\begin{align*}
\mu & \quad \text{Blood viscosity} \quad 4.00 \times 10^{-3} \text{ Pa.s} \\
P_{in} & \quad \text{SA inlet blood pressure} \quad 2506 \text{ Pa (18.8 mmHg)} \\
P_{out} & \quad \text{DV outlet blood pressure} \quad 0.147 \text{ Pa (1.10 } \times 10^{-3} \text{ mmHg)} \\
\kappa_{\text{empty}} & \quad \text{Maximum hydraulic conductivity} \quad 0.52 \text{ mm}^2
\end{align*}
\]

References: [12, 55, 57, 182]
Chapter 3. Predicting maternal blood flow in a placental subunit

Figure 3.4: Streamlines predicted for a placental unit under baseline conditions. Maternal blood entering from the SA into the IVS follows irregular streamlines that have a tendency to pass through villous tissue with higher hydraulic conductivity before draining through the DVs.

model from Chernyavsky et al. [55] and experimental [104] studies which describe high flow and pressure near the SA inlets and a rapid dissipation of flow and pressure in regions comprising dense villous tissue, where blood percolates rather than streams. However, unlike the regular streamlines which conform to the hemispherical domain in an earlier porous medium model due to the placement of DVs with respect to the SA [55], the current model generates irregular streamlines which have a tendency to pass through regions with higher hydraulic conductivity, which is expected for a medium with spatially varying porosity. These regions with higher hydraulic conductivity are made up of non-terminal villous branches with sparse terminal tissue blocks (TBs).

Villus density

Figure 3.5 shows the effect of varying terminal villus density ($\phi_{TV}$) on the flow velocity emerging from the SA. Consistent with previous studies [55, 56], our model demonstrates an increased impedance to maternal blood with the increase in the terminal villus density.

The effects of a central cavity

For consistency with previous models [53, 55], the model was solved using an artificially applied ‘central cavity’, represented by a relatively villus-free region with a hydraulic conductivity of $\kappa_{\text{empty}}$, having the same length and width of a typical cavity. Here, we assumed that the central cavities are completely filled by maternal ‘jets’ emerging from the SAs into the IVS and adopted an estimated ‘jet’ width of 4 mm and length ranging
3.3. Results

**Figure 3.5:** The effects of terminal villus density ($\phi_{TV}$) on IVS flow. Flow velocity decreases due to increased impedance from denser terminal villous tissue.

**Figure 3.6:** The effects of central cavities (CC) on IVS flow. (a) Flow streamlines show that maternal blood projects deeper towards the chorionic plate in the presence of a central cavity. (b) Flow velocity emerging from the SA increases with the length of the central cavity.

(from 3.0–8.6 mm (as measured using colour Doppler ultrasonography [123] for a placenta at 33 weeks) as the dimension of the cavity in our model.

In the presence of a central cavity, the predicted maternal blood flow patterns are consistent with the ‘physiological concepts’ hypothesised by Ramsey et al. [128] and maternal blood is clearly shown to project towards the chorionic plate due to the velocity of blood, resulting in delivery of maternal blood to deeper parts of the placental tissue.)
Chapter 3. Predicting maternal blood flow in a placental subunit

(Figure 3.6a). When the length of the central cavity is increased, Figure 3.6b shows that maternal blood enters into the IVS at higher speed.

3.3.2 The branching structure of non-terminal villi

Length of villous elements ($l_s$)

Often, pathology is associated with a change in the balance of vessel branching and elongation [143, 187]. While the villous tree ‘fills’ the placental volume, it may do so with fewer, longer branches (or conversely more, shorter branches). By changing $l_s$, trees with these branching properties were generated. A range of $l_s > 1.6$ mm was considered, as below this range the generated tree does not fill the thickness of the term placenta. Figure 3.7 shows the effects of $l_s$ on the velocity of blood emerging from the SA. As $l_s$ increases from 1.6 mm, the villous tree begins to fill more of the IVS. With an increase in flow impedance, flow velocity into the IVS decreases. However, as $l_s$ increases beyond $l_s = 2.4$ mm, the spaces between branches increases to a point where it is sparse enough that blood flow velocity into the IVS increases again under the same driving pressure. As shown in Figure 3.8, the increase in spaces between branches also gives rise to flow paths with lower flow resistance between the SA and DVs, resulting in flow streamlines that do not penetrate as deep into the placental tissue, as demonstrated by the streamlines generated by the tree with $l_s = 3$ mm.

Branch angle ($\theta_b$)

Figure 3.9 shows the effects of varying $\theta_b$ (while holding all other parameters constant) on the velocity of blood emerging from the SA. Inlet flow velocity varies non-linearly with $\theta_b$ with a peak at $\theta_b = 24^\circ$. The different branch angles give rise to different conductivity fields. As illustrated in Figure 3.10, villous branches and TBs spread out more around the inlet as $\theta_b$ increases, thereby allowing maternal blood to flow more freely into the IVS under the same driving pressure, resulting in an increase in inlet flow velocity. However, at some point ($\theta_b \geq 26^\circ$) the gaps between the tree branches become so big that highly conductive paths are formed and flow is shunted directly from the SA to the DVs, bypassing the depth of villous tissue.
3.3. Results

Figure 3.7: The effect of the length of the stem branch \((l_s)\) on IVS flow. The dotted line indicates the nominal value of \(l_s\). While all other parameters were held constant, increasing \(l_s\) effectively increases the length of all branches in the villous tree. Blood flow velocity into the IVS is non-linear with \(l_s\). Inlet flow velocity decreases as \(l_s\) increases initially due to the increased impedance by longer branches. Beyond \(l_s = 2.4\) mm, the spaces between branches become so sparse that maternal blood flow starts to increase.

Figure 3.8: Streamlines predicted for villous trees with \(l_s = 2\) mm and \(l_s = 3\) mm. Fewer, longer villous branches and fewer terminal villi fit into the domain with the increase in \(l_s\). Trees with higher \(l_s\) provide less impedance to maternal blood flow due to the sparse spaces between its villous branches. These spaces also create conductive flow paths, resulting in bypass of maternal flow from the SA to DVs without deep penetration of the placental thickness.
Chapter 3. Predicting maternal blood flow in a placental subunit

Figure 3.9: The effects of branch angle ($\theta_b$) on IVS flow. The dotted line indicates the nominal value of $\theta_b$. Velocity of maternal emerging from the SA varies non-linearly with $\theta_b$ with a peak at $\theta_b = 24^\circ$.

Figure 3.10: Streamlines predicted for villous trees with $\theta_b = 18^\circ$, $\theta_b = 24^\circ$ and $\theta_b = 26^\circ$. Regions around the inlet are less filled with villous tissue with increasing $\theta_b$, giving rise to deeper flow penetration into the placental tissue. Beyond $\theta_b = 26^\circ$, the large spacings between branches generate paths of high hydraulic conductivity between the inlet and outlets, resulting in arteriovenous shunting.

Number of branching generations ($n_b$)

$n_b$ was varied while holding all other parameters constant. With less than 11 branching generations, the tree does not cover the thickness of the term placenta, which is not physiological, and so only $n_b$ between 11 and 17 were considered. The effects of $n_b$ on the velocity of blood emerging from the SA are shown in Figure 3.11. This parameter has only a small effect on flow velocity in the absence of concurrent changes in branch length or angle. This is because the orientation of villous branches and TBs near the SA inlet remain similar even as $n_b$ is increased as illustrated in Figure 3.12. This results in similar tissue resistance to inlet flow and hence, only small effects on inlet velocity are observed when $n_b$ is varied.
3.3. Results

Figure 3.11: The effects of the number of branching generations \( (n_b) \) on IVS flow. The dotted line indicates the nominal value of \( n_b \). A range between 11 and 17 branching generations was considered, below this range the generated tree does not fill the thickness of the term placenta. Blood flow into the IVS remains almost constant as \( n_b \) was varied.

Figure 3.12: Flow streamlines for villous trees with \( n_b = 12 \) and \( n_b = 15 \). Villous branches and TBs are oriented similarly near the SA for both cases, providing similar resistance to inlet flow even though the streamline patterns are different.

Random variability and asymmetry

An asymmetric villous tree with random branch angles between 10° and 40° was generated to evaluate the influence of random variability of villous tree structure on model predictions. With the introduction of asymmetry, a velocity boundary condition was applied at the SA to obtain \( Q_m \) of 83.3 mm³/s (5 ml/min) as pressure boundary conditions at the inlet SA and outlet DVs are non-applicable. The flow profile (Figure 3.13) differs from the baseline case and follows an asymmetric pattern in accordance with the conductivity field generated based on the properties of the villous tree.
Chapter 3. Predicting maternal blood flow in a placental subunit

Figure 3.13: Streamlines predicted for a placental subunit containing an asymmetric villous tree with random branch angles. The streamlines follow an asymmetric pattern that corresponds to the more conductive path directed by the tree properties.

3.4 Discussion

In this chapter, we have presented a model of maternal blood flow in the IVS of a placental subunit, which incorporates key geometric features of villous tree architecture. While computational models of placental blood flow have typically adopted a simplified homogeneous geometry for the IVS [55, 56], the current model attempts to capture the different structural characteristics of the villous tree from the stem villi to terminal villi level to provide a representative anatomically based description of how placental structures influence the maternal blood flow around them.

Consistency with previous studies

The predicted IVS blood flow pattern resulting from the baseline case is consistent with the ‘physiological concept’ proposed in cine-angiography and injection studies [128], whereby it was postulated that streams or ‘jets’ of maternal blood enter the IVS and percolate radially through the densely packed villous tree due to dissipation of flow and pressure. Flow behaviours predicted by the model with structural variations of terminal villus density and introduction of a central cavity are also consistent with a previous model that assumed a uniform porous medium to represent the placenta [169].

The effects of villous branches

The structure of the branching component of the villous tree (the stem and intermediate villi) plays an important role in channelling blood flow in the IVS. As branches of the villous tree act as a barrier to blood flow, and empty spaces between the branches allow
free movement of maternal blood, the distribution of villous branches within the IVS influences their contribution to IVS conductivity. In addition, the branching structure of the villous tree also determines how the villous tree fills out the IVS, which could influence the number and distribution of terminal villi (which also contributes to IVS conductivity) due to space limitation in the IVS. Using the trees generated under baseline conditions and asymmetrical conditions, we demonstrated irregular flow streamlines which are shaped by the varying conductivity field as maternal blood tends to traverse regions of non-terminal villous branches with sparser TBs over regions with dense TBs. The branch angle, length and diameter of the stem and intermediate villi, all influence how villous tree branches are spread out and fill the space in the placenta. Stem and intermediate villi with shorter lengths or smaller branch angles are more closely packed and more resistant to maternal blood flow while longer villi or villi with bigger branch angles are more spaced out, which allows maternal blood flow to penetrate faster and further through the IVS.

The distribution of villous tissue around the SA inlet is especially important as it determines the tissue impedance to incoming flow, which in turn affects flow velocity and the path taken by the flow in the IVS. If the villous branches are too sparse, maternal blood will follow the path of least resistance and flow through the gaps between the villous branches and drain directly from the SA into the DVs, without penetrating deeper into the IVS. Such arteriovenous shunting in the IVS is disadvantageous for material exchange, which may also explain why villus-free margins between villous trees and the basal plate have never been observed or reported. Although previous models have suggested that shunts can arise when DVs are close to the SA [55] or when highly permeable regions are created by villi distortion due to high speed flow near the inlets and outlets [53], our model introduces a third mechanism whereby shunts can arise due to the branching structure of the villous tree.

Model limitations

There are several studies which aim to quantify the branching properties of villous structures [43, 186], and these studies are becoming increasingly quantitative allowing incorporation of branching and 3D rotation angles [186]. Our branching villous model aims to match as closely as possible to these existing studies, in terms of measured tree properties including branch numbers and lengths. With advancements in imaging techniques, anatomical studies using high resolution images (for example, micro-CT) of the villous tree geometry can be incorporated to improve model accuracy.
Like other models in the literature \cite{53, 55, 56, 170}, the model follows the idealised configuration of one SA and two DVs to a single villous tree. However, as discussed in Section 2.3.2, this idealised configuration is typically observed in the peripheral regions of the placenta, and rarely occurs in the central region where villous trees are bigger \cite{12}. Given the dependence of maternal blood flow in the IVS on the location of SAs \cite{55} and the orientation of villous tree structure to SA openings, we will establish a modelling framework in Chapter 6 that better represents the anatomical distribution of SAs of a human placenta for future investigation of this placental structure-function relationship.

In line with the 2D simplification of the 3D villous tree structure, we have modified the Kozeny-Carman formula (which is typically applied to model 3D media) to use the area fraction occupied by the villous tree instead of volume fraction to determine a spatially varying conductivity field for approximating IVS flow in 2D like previous models \cite{53, 170}. Even though such consideration may not completely reflect the 3D villous geometry and flow dynamics in the placenta, it is sufficient for preliminary identification of parameters with major influence on placental function to motivate further extension of the model into 3D as explored in Chapters 4 and 6.

Although our model assumed that the IVS is a porous medium and approximated blood flow in the IVS using Darcy’s law as in previous models, our model moved away from a uniformly porous medium and took a step further by using a sampling grid to capture the regional variation in porosity based on the structure of the villous tree. In our model, a sampling grid window of 2 mm was used to capture regional variations in porosity while satisfying the assumption that the IVS is a porous medium. This is consistent with the findings of Chernyavsky et al. \cite{169} in which villous structure in a 2D section is considered as a homogenised or slowly varying porous medium if the sampling grid window size is bigger than 1 mm. Even though refinement of the sampling grid will affect the porosity distribution and give rise to a distribution that approaches the geometry of the villous tree, model solution may become unreliable since Darcy’s law is no longer suitable for approximating flow given that the assumption of the IVS as a porous medium loses its validity. Ideally, the villous tree should be represented with all its stem and intermediate villus branches as well as its terminal convolutes, and maternal flow should be simulated based on such detailed structures. However, given the complexities involved in representing the convoluted structure of terminal villi, we took a simplified approach of homogenising the ‘random’ structure of terminal villi into a tissue block with an
isotropic conductivity. While it is possible to account for conductivity anisotropy or even solve for Navier-Stokes flow around the villous tree as demonstrated by Lecarpentier et al. \cite{170}, computational cost remains an issue with such a complicated geometry, especially if flow is to be modelled on a whole organ level.

### 3.5 Summary and conclusions

In this chapter, we described a maternal blood flow model that accounts for villous tree geometry. By varying the structure of the villous tree, the model demonstrates how villous tissue contributes to IVS conductivity and predicts how maternal blood flow in a human placental subunit is influenced by the non-uniform conductivity distribution arising from variations in the villous structure. This model also introduced a new mechanism where arteriovenous shunts in the placenta can arise from the branching structure of the villous tree.

In Chapter 4, we will introduce fetal blood flow into our model, and apply the model to examine how maternal and fetal blood flow in the placenta affect the generation of PGSE signals. We will use this model to assess the applicability of the IVIM technique for blood flow visualisation in the human placenta.
Chapter 4

Interpreting magnetic resonance images of placental blood flow

4.1 Motivation

Imaging the spatial distribution of placental perfusion is not straightforward. As such, diagnosis of abnormal placental resistance is normally inferred from 2D Doppler ultrasonography [188]. The gold-standards for imaging perfusion distributions use ionising radiation imaging and contrast agents (e.g. X-ray computed tomography and positron emission tomography), both of which are generally contraindicated in pregnancy [189]. However, these issues are avoided in ultrasound and MRI. Ultrasound is able to produce measures of perfusion including vascularisation indices and flow indices [190], but their relationship to actual flow and tissue properties is uncertain [191, 192]. Ultrasound is limited by the penetration depth of the ultrasound beam, which makes it challenging to reliably image the placenta if it is in a posterior location, or if the transducer is obstructed by tissues (like in obesity) [193]. The field of view is also limited, so visualisation of the entire placenta is difficult as the size of the placenta increases over gestation [193]. On the other hand, MRI of the placenta allows a greater tissue contrast than ultrasound and a wider field of view for functional imaging [193, 194].

A promising approach for analysing placental blood flow without the use of contrast agents is to employ the pulsed gradient spin echo (PGSE) sequence and the intravoxel incoherent motion (IVIM) technique. The IVIM method has been used successfully to
quantify changes in vascularisation between population groups in the brain, renal tissue, and other vascularised tissues [195]. IVIM has been applied to understand the nature of placental perfusion [196, 197], and has shown differences in the perfusion heterogeneity between normal and growth restricted pregnancies [197].

The IVIM method was developed based on a mathematical model of the magnetisation of tissue following the PGSE signal. The model assumes that perfusion of tissue (whatever the type of tissue) is a quasi-diffusion process, as illustrated in Figure 4.1a. Essentially, the assumption relies on a vascularised tissue approximating a network of randomly oriented tubes. This means that blood flow through the tubes is close to random, and so the governing equations for magnetisation in this network can be decomposed into two diffusion processes, that of magnetisation in stationary tissue, and that of magnetisation in the moving blood [195, 198, 199].

Unlike other tissues where the IVIM method has been applied to a micro-circulatory network embedded within the tissue (Figure 4.1a), imaging of the placenta must take into account its unique structure at the interface of two independent blood circulations (maternal and fetal, Figure 4.1b). Even if we assume that the fetal circulation in the placenta behaves in a quasi-diffusive manner as it resides in the complex villous tree structure, it is unclear whether the maternal circulation is also quasi-diffusive, and if it has the same properties as the fetal circulation. Conventionally, analysis of IVIM images is broken down to a biexponential fit to signal data under different imaging protocols by assuming a vascularised network is embedded in stationary tissue [195]. This fit approximates two apparent diffusion coefficients: $D$, representing the diffusion of water molecules in stationary tissue, and $D^*$, representing the apparent diffusion of moving blood. In the placenta, it is unknown whether a single $D^*$ representing all moving blood (fetal and maternal) is sufficient, and indeed $D^*$ has been found to be subjected to high fitting errors in placental IVIM [196]. Also, there are potentially some regions within the placenta where flow is non-uniform within a voxel but cannot be approximated as a quasi-diffusion process, for example, close to the openings of the maternal arteries. It remains unknown how PGSE signal is affected by such blood pattern and if classical assumptions made in the IVIM method are applicable to such flow.

To investigate these issues, we introduce a component of fetal blood flow to the maternal blood flow model described in Chapter 3, and present a mathematical model that
4.2 Theoretical background

Before modelling the influence of placental structure on IVIM signal, a knowledge of nuclear magnetic resonance (NMR) theory is required. Here, we introduce important concepts, including how signals are generated and how the IVIM method was developed.

4.2.1 Nuclear spin and magnetic fields

Using the principles of NMR, MRI allows visualisation of internal organs by using magnetic fields to influence the spin of atoms in the tissues of the body. Spin is an
intrinsic property of atoms with an odd number of protons, neutrons or both; atoms with an even number of protons, neutrons or both do not possess an intrinsic spin. In clinical imaging, signals generated from the hydrogen atoms are measured as they are the most abundant isotope in the human body, since they are found in fats and water.

Nuclear spin is quantified by the spin quantum number, \( I \), which is a half-integer if the total number of protons and neutrons is odd, and an integer if the total number of protons and neutrons is even. \( I \) is used to calculate the magnitude of the total intrinsic angular momentum of the nucleus, \( I \), defined by

\[
|I| = \frac{h}{2\pi} \sqrt{I(I + 1)},
\]

(4.1)

where \( h \) is Planck’s constant \((6.63 \times 10^{-34} \text{ m}^2\text{kg/s})\). The magnetic dipole moment of the nucleus, \( \mu_n \), is given by [200]

\[
\mu_n = \gamma I,
\]

(4.2)

where \( \gamma \) is the gyromagnetic ratio of the nucleus, which is unique for each nucleus and is calculated using

\[
\gamma = \frac{q_n}{2m_n} g_f,
\]

(4.3)

where \( q_n \) is the charge, \( m_n \) is the mass and \( g_f \) is the dimensionless magnetic moment (g-factor) of the nucleus (for hydrogen, \( \gamma = 2.68 \times 10^8 \text{ rads}^{-1}\text{T}^{-1}(42.58 \text{ MHzT}^{-1}) \) [201]).

Nuclear spins are randomly oriented in an unmagnetised state, and align like tiny compass needles when subjected to an external magnetic field, \( \mathbf{B} \). Locally, the magnetic dipole moment of the nucleus experiences a torque, \( \mathbf{\tau_n} \), given by

\[
\mathbf{\tau_n} = \mathbf{\mu_n} \times \mathbf{B},
\]

(4.4)

which aligns the magnetic moment with the magnetic field [200]. Note that a nucleus needs to have spin in order to be sensitive to a magnetic field (as is applied in MRI), otherwise \( \mu_n = \tau_n = 0 \).

As a result of torque, the magnetic moment precesses around the axis of the applied magnetic field (or wobbles like a spinning top). This is known as the gyroscope ef-
fect and the resonant angular frequency of the nuclear spin is known as the Larmor frequency, $\omega_L$ [200], with

$$\omega_L = \gamma B.$$  \hspace{1cm} (4.5)

To obtain an NMR signal, an electromagnetic radiofrequency (RF) pulse is applied, and energy from the RF pulse is absorbed by the spins which causes them to be in phase with each other and precess at their Larmor frequency. Upon removal of the RF pulse, an NMR signal is generated as the absorbed energy is released when the excited spins which were aligned in the direction of the applied magnetic field begin to dephase (T2 relaxation) and return to their original orientation (T1 recovery).

The net magnetisation of the magnetic moment, $\mathbf{M} = M_x \hat{i} + M_y \hat{j} + M_z \hat{k}$, in the presence of the externally applied magnetic field is [200]

$$\frac{d\mathbf{M}}{dt} = \omega_L \times \mathbf{M}. \hspace{1cm} (4.6)$$

This equation describes overall magnetisation rather than the motions of individual nuclear moments. Equation 4.6 was modified by Bloch [200] to factor in the effects of spin excitation and relaxation on the macroscopic behaviour of $\mathbf{M}$ due to an RF pulse. Assuming a net magnetisation of $M_0$ along the $z$-axis at equilibrium, application of the RF pulse causes the spins to precess in the transverse $x$-$y$ plane. Magnetisation is hence represented by the Bloch equation as

$$\frac{d\mathbf{M}}{dt} = \gamma \mathbf{M} \times \mathbf{B} - \frac{M_x \hat{i} + M_y \hat{j}}{T_2} - \frac{(M_z - M_0)\hat{k}}{T_1}, \hspace{1cm} (4.7)$$

where T2 relaxation occurs in the $x$-$y$ plane at a rate of $1/T_2$ and T1 recovery occurs along the $z$-axis at a rate of $1/T_1$.

### 4.2.2 Pulsed Gradient Spin Echo (PGSE)

PGSE is a technique used in MRI for generating images with diffusion weighted contrast. The PGSE sequence involves implementing a pair of $\pi/2$ rad and $\pi$ rad RF pulses to create a spin echo, as shown in Figure 4.2. Gradient pulses of duration $\delta$ and magnitude $G$ are applied along the $z$-axis before and after the $\pi$ rad pulse. The first gradient
Figure 4.2: Schematic representation of the PGSE sequence. A pair of $\pi/2$ rad and $\pi$ rad RF pulses are applied with equal gradient pulses of magnitude $G$ on either side of the $\pi$ rad RF pulse, each lasting $\delta$. The first gradient pulse is applied $t_1$ after the start of the PGSE sequence and the second gradient pulse is applied $\Delta$ after the first gradient pulse. Signal generated by the PGSE sequence is acquired at echo time ($TE$).

Figure 4.3 illustrates how an ensemble of spins behave in response to the different pulses implemented during a PGSE sequence for fluid undergoing different types of motion. At the start of the sequence, nuclear spins are aligned along the magnetic field $B_0$ and possess a net magnetisation ($M_0$) along the $z$-axis before the application of a $\pi/2$ rad RF pulse. With the introduction of a $\pi/2$ rad RF pulse, the macroscopic magnetisation is flipped from the $z$-axis to the $x$-$y$ plane. As the $\pi/2$ rad RF pulse is switched off, the spins attempt to revert to their equilibrium state through loss of the energy gained from the RF pulse and they dephase to realign themselves with the $B_0$ field. When a $\pi$ rad RF pulse is applied at time $TE/2$, the spins are reversed by twice the amount they have moved since $t = 0$. By thinking of the spins as hands of a clock, this spin reversal essentially cancels the response of the spins induced by the $\pi/2$ rad RF pulse if the spins are not influenced by any form of fluid movement. The spins revert to their original orientation by signal acquisition time $TE$, and there is essentially no signal attenuation (i.e. $S(TE) = S(0)$) (Figure 4.3a). However, if dephasing of the spins is confounded by atomic diffusion before the application of the $\pi$ rad RF pulse, the spins are unable to revert back to their original starting positions by signal acquisition time.
and the degree of dephasing varies within the material being imaged \cite{202} (Figure 4.3b), resulting in a detectable spin-echo amplitude attenuation. In the presence of coherent flow without any atomic diffusion, outflowing spins are constantly being replaced by incoming spins. As shown in Figure 4.3c, the spins return to being in phase by signal acquisition time \cite{202}. However, they experience a net phase shift which is proportional to the velocity of flow ($v$), resulting in a signal represented by

$$S(TE) = S(0)e^{i\gamma G \Delta v}.$$  \hspace{1cm} (4.8)

Mathematically, magnetisation throughout the PGSE sequence is described by a modified Bloch equation which accounts for how magnetisation of the spins are altered by both fluid movement and diffusion \cite{198}

$$\frac{\partial M}{\partial t} = \gamma M \times B - \frac{M_x \hat{i} + M_y \hat{j}}{T_2} - \frac{(M_z - M_0)\hat{k}}{T_1} + D \nabla^2 M - \mathbf{v} \cdot \nabla M,$$  \hspace{1cm} (4.9)

where $D$ is the diffusion coefficient and $\mathbf{v}$ is the velocity of fluid movement. The magnetic field, $B$, consists of the static magnetic field ($B_0$) applied along the $z$-axis superimposed with a spatially varying magnetic gradient field ($G_z$), which is also applied along the $z$-axis:

$$B = (B_0 + G_z)\hat{k},$$  \hspace{1cm} (4.10)

where $G$ is the magnitude of the gradient pulses applied during the PGSE sequence. As the change in magnetisation in the transverse $x$-$y$ plane provides the NMR signal, we consider transverse magnetisation, $m = M_x + iM_y$, to reduce Equation 4.9 to

$$\frac{\partial m}{\partial t} = -i\omega_0 m - i\gamma (Gz)m - \frac{m}{T_2} + D \nabla^2 m - \mathbf{v} \cdot \nabla m,$$  \hspace{1cm} (4.11)

where $\omega_0 = \gamma B_0$. Additional detail on the derivation of Equation 4.11 from Equation 4.9 is provided in Appendix B.1.

Since $m$ can be decomposed into a component that is unaffected by T2 relaxation ($\psi$) and a component that is affected by T2 relaxation ($e^{-i\omega_0 t - t/T_2}$) \cite{198}:

$$m = \psi e^{-i\omega_0 t - t/T_2},$$  \hspace{1cm} (4.12)
Figure 4.3: Nuclear spins at different stages of the PGSE pulse sequence. Spins are in phase and aligned on the $x$-$y$ plane at the start of the PGSE sequence ($\pi/2$ rad RF pulse). The spins start to dephase and return back to its equilibrium positions after the $\pi/2$ rad RF pulse is switched off (before $\pi$ rad RF pulse). Application of a $\pi$ rad RF pulse reverses the spins by twice the amount they moved since the start of the PGSE sequence (after $\pi$ rad RF pulse). (a) For spins in a stationary fluid, the $\pi$ rad RF pulse effectively cancels out the spins movement caused by the $\pi/2$ rad RF pulse and all spins revert back to their original starting positions by $t = TE$, thereby generating a signal with the same intensity as the signal at the start of the PGSE sequence. (b) Under the influence of atomic diffusion, dephasing is confounded by random walk of the spins and each spin experiences a different degree of dephasing. This results in the inability of the spins to revert to its original orientation by $t = TE$ and hence, signal is attenuated. (c) Spins undergoing coherent flow are constantly replaced by spins in the incoming flow. These spins experience a net phase shift but they revert to being in phase by $t = TE$. 
substitution of Equation 4.12 into Equation 4.11 results in

$$\frac{\partial \psi}{\partial t} = -i\gamma(Gz)\psi + D\nabla^2\psi - \mathbf{v} \cdot \nabla \psi.$$  (4.13)

Equation 4.13 solved over the course of the PGSE sequence generates a solution for \(\psi\) in the form of \(\psi = Ae^{-i\varphi}\), where \(A\) is the magnitude of \(\psi\), and \(\varphi\) is the phase of \(\psi\) \((\varphi = \gamma Gzt)\). The total signal strength from an imaging voxel is determined by integrating the magnetisation of the population of spin phases in the voxel space:

$$S = \int Ae^{-i\varphi}e^{-i\omega_0t/T_z}dV,$$  (4.14)

where \(V\) is the volume of a voxel. Therefore, signal attenuation due to fluid movement and diffusion is obtained by normalising the signal intensity at acquisition time by the signal at the start of the PGSE sequence.

When an imaging voxel contains fluid movement by diffusion only \((v=0)\), Equation 4.13 can be solved analytically. The signal at the end of the PGSE sequence is constant for any given diffusion sensitivity parameter \(b\) and the relationship between the signal amplitude \(S\) and \(b\) is \([198, 199]\)

$$S(b) = S(0)e^{-bD},$$  (4.15)

where \(D\) is the diffusion coefficient of fluid molecules, and

$$b = \gamma^2G^2\delta^2\left(\Delta - \frac{\delta}{3}\right).$$  (4.16)

By altering \(G\) (and so \(b\)), a range of signal magnitudes is measured and the diffusion coefficient \(D\) is estimated via an exponential fit to signal data on a voxel-by-voxel basis.

In vascularised tissue where blood flows in randomly oriented capillaries, flow is typically conceptualised as a quasi-diffusion process with a diffusion coefficient \(D^*\) and Le Bihan et al. [195] demonstrated that Equation 4.15 is modified to

$$S(b) = S(0)\left[(1-f)e^{-bD} + fe^{-bD^*}\right],$$  (4.17)
where $f$ is the fraction of moving blood in an image pixel. For data acquired across a range of $b$ values, a biexponential fit is used to determine $D$ and $D^*$, with $D^*$ and $D$ influencing $S$ predominantly at low and high values of $b$, respectively [203].

### 4.3 Methods

Here, we solve the classical magnetisation theory (the Bloch equation, Equation 4.13) numerically under the PGSE protocol using the model of placental blood flow as described below. This allows us to investigate whether flow patterns derived from placental structures behave in a quasi-diffusive manner and to predict signal attenuation due to flow profiles that might be seen in the placenta.

#### 4.3.1 Model geometry

Simulations of PGSE signal were conducted in a geometry reflective of an image ‘slice’ that sits within the placental boundary. As a basis for modelling flow profiles that are representative of placental blood flow, we constructed a 3D hexahedral domain with thickness $\tau$ and width $\epsilon$ to represent a placental subunit comprising a fetal blood circulation, and a maternal circulation which is supplied through a central SA and drained by two DVs. To gain a better understanding of how blood flow in the placenta contributes to signal attenuation, we used five different models as shown in Figure 4.4 to examine the individual behaviour of the fetal and maternal circulations as well as their combined effects on PGSE signal generation.

**Model 1–Fetal blood flow in randomly oriented network of vessels**

The theory developed to analyse IVIM signals assumes a quasi-random flow profile [195]. Using a model composed of vessels that are connected to form a random path, we aim to verify our model framework. For the first vessel segment, five random numbers were generated using a random number generator (the RAND function embedded in Matlab), to define the midpoint of the vessel (its $x$-, $y$-, and $z$-coordinates), the angle between the longitudinal axis of the vessel and the $z$-axis ($0 < \theta < \pi$), and the angle of rotation about the $x$-$y$ plane ($0 < \sigma < 2\pi$). The first vessel was assigned a radius $r$ and length $l$. The end coordinate of the vessel was used as a starting point for the next vessel segment, and new values for $\theta$ and $\sigma$ were generated using the random number generator. The
4.3. Methods

Figure 4.4: Models used to investigate signal attenuation by placental blood flow. Only fetal blood flow was simulated in Models 1 and 2. Fetal vessels were represented as a randomly oriented network of vessels in Model 1, and as a tree-like structure in Model 2. Only maternal blood flow was simulated in Models 3 and 4. Maternal blood flow was simulated as flow in a uniform porous medium in Model 3, while Model 4 simulated maternal blood flow in a porosity field that varies spatially with the structure of a villous tree. Model 5 simulated complete perfusion within a placental subunit by combining fetal perfusion in a vessel tree (Model 2), molecular diffusion in villous tissue, and maternal circulation in the IVS (Model 4).

Model 2–Fetal blood flow in villous vessel tree structure

No attempt has been made in the literature to capture the nature of PGSE signal generated from flow in a tree-like structure. In order to verify whether flow in a villous vessel tree behaves in a quasi-diffusive manner, we generated a villous tree structure in 3D using the method described in Section 3.2.1. Here we assumed that the fetal vascular tree follows the same course of branching as the generated villous tree structure, and constructed a fetal arterial tree by adjusting the branch diameters of the generated villous tree structure to reflect the diameter of fetal vessels. The diameter of villus
artery \( (d_A) \) in the trunk of the villous tree was derived from measurements presented in the literature \([204]\), while diameter of subsequent blood vessels were controlled using the Strahler diameter ratio \( (D_{SR}) \), which was fitted to ensure that the diameter of the smallest vessel is at least the size of a capillary. \( D_{SR} \) was also fitted to generate a total fetal vascular volume that matches the volume estimated using stereological methods \([42, 205]\). To obtain the total fetal vascular volume, we assumed that the fetal venous tree follows the arterial tree, with each venous vessel \( (d_v) \) having twice the diameter of its associated artery \([206]\). In this model, we only examined how spin magnetisation and signal generation are affected by the structure of the fetal arterial tree since we do not anticipate additional insights with the inclusion of the venous tree.

Model 3–Maternal blood flow in a uniform porous medium

Following earlier studies \([53, 55]\), the 3D domain was assumed to be a porous medium with a uniform hydraulic conductivity of \( \kappa_{\text{uniform}} \). The model was applied to investigate how local variations in the flow magnitude and direction of maternal blood flow in the IVS affect spin magnetisation and hence generation of PGSE signals.

Model 4–Maternal blood flow in a porous medium with hydraulic conductivity as function of tree structure

In this model, the porosity of the domain was varied spatially with respect to the representative villous tree structure as described in Section 3.2.2. Comparison of results from this model and Model 3 (uniform porosity) will inform how flow in the IVS, and the nature of PGSE signals, are affected by the structure of villous trees.

Model 5–Fetal and maternal blood flow in full placental blood flow model

We combined (1) maternal blood circulation modelled as flow in a non-uniform porous medium with hydraulic conductivity varying as a function of the villous tree structure (Model 4), (2) molecular diffusion in villous tree tissue, and (3) fetal perfusion in vessels embedded in the villous tree (Model 2) to examine the nature of signal generated by the placental structure and the physiological significance of the resultant IVIM parameters.
4.3 Methods

4.3.2 Fetal blood flow

**Governing equations and solution procedure**

A uniform flow velocity $v$ was implemented across all vessel segments in Model 1. For Model 2, to simulate steady-state fetal blood flow in the fetal vascular tree, each blood vessel was represented in the model as a rigid tube defined by a vector between the start and end point of the vessel. The resistance of each vessel ($R_v$) was calculated using

$$R_v = \frac{8\mu l}{\pi r^4},$$  

(4.18)

where $l$ is the vessel length, $r$ is the vessel radius, and $\mu$ is the viscosity of blood.

The pressure drop ($\Delta P_v$) across each vessel is the product of the volumetric blood flow rate through the vessel ($Q_v$) and the resistance of the vessel, which is expressed as

$$\Delta P_v = Q_v R_v.$$  

(4.19)

By applying conservation of flow, blood flow through a parent vessel ($Q_p$) was distributed into the two daughter vessels ($Q_{d1}$ and $Q_{d2}$) at each bifurcation using

$$Q_p = Q_{d1} + Q_{d2}.$$  

(4.20)

With the assumption of pressure continuity across each bifurcation, where the pressure at the outlet of a parent branch is defined to be equal to the pressure at the inlet of its daughters, we derived a system of linear equations to be solved for the unknown pressures at each bifurcation and volumetric flow in each branch.

**Boundary conditions**

In order to solve for pressure and flow distribution through the fetal arterial geometry, boundary conditions were defined with volumetric flow at the inlet ($Q_{in}^f$) and a constant pressure ($P_t$) for all terminal vessels.

4.3.3 Modelling the PGSE sequence

**Governing equations**

Spin magnetisation was modelled with Equation 4.13. The PGSE sequence was modelled
in its entirety by dividing the sequence into five time intervals (Figure 4.2)

\[
\frac{\partial \psi}{\partial t} = \begin{cases} 
D \nabla^2 \psi - \mathbf{v} \cdot \nabla \psi, & t \in [0, t_1) \cup [t_1 + \delta, t_1 + \Delta) \cup [t_1 + \Delta + \delta, TE], \\
-i\gamma (G_z) \psi + D \nabla^2 \psi - \mathbf{v} \cdot \nabla \psi, & t \in [t_1, t_1 + \delta) \cup [t_1 + \Delta, t_1 + \Delta + \delta].
\end{cases}
\tag{4.21}
\]

As the maternal circulation is independent of the fetal circulation, Equation 4.21 was solved separately for each circulatory system with \(D\) equivalent to the diffusion coefficient of water \((D = 2.5 \times 10^{-3} \text{ mm}^2/\text{s} [207])\) and \(\mathbf{v}\) as defined by the respective flow fields derived from maternal blood flow (Section 3.2.2) and fetal blood flow models.

**Initial and boundary conditions**

An initial \(\psi\) was assigned throughout the domain to represent the alignment of spin phases at the start of the sequence, \(\psi(x, y, z) = 1\). At time \(TE/2\), application of the \(\pi\) rad pulse sets the phase of the precessing magnetisation \((\phi^*)\) at each point back by twice the angle it has precessed, which is equivalent to

\[
\phi(x, y, z, TE/2) = -\phi^*(x, y, z, TE/2).
\tag{4.22}
\]

As the magnetisation state of spins entering into the solution domain is unknown, different boundary conditions at the blood flow inlet were tested (Appendix B.2.2). \(\psi\) was fixed with a magnitude \(A = 1\) and a phase \(\phi = 0\) at the inlet (i.e. magnetised blood enters the domain).

**Finite element mesh and solution procedure**

A Lagrange-Galerkin finite element solver was developed to solve Equation 4.21 (Appendix C) [208]. This is a split operator approach in which, over each time step, the departure point of blood at each mesh node was calculated and \(\psi\) at that mesh node was initiated with the spin magnetisation at the blood’s departure point at the start of the timestep. The updated magnetisation field was then used as an initial condition to calculate the new magnetisation distribution resulting from diffusion using the Galerkin finite element method.

A finite element mesh comprising linear elements was used. For spatial convergence, an element size of 0.04 mm was used to model spin magnetisation in the
1D flow occurring in the fetal blood flow model (Appendix B.2.3), while an element size of $0.5 \text{ mm} \times 0.5 \text{ mm} \times 0.5 \text{ mm}$ was used to model magnetisation in the maternal blood flow model. A timestep of $1 \times 10^{-4} \text{ s}$ was used to achieve temporal convergence (Appendix B.2.3).

### 4.3.4 Post-processing and model outputs

**Signal generation**

The typical size of an image voxel for this imaging modality is $3.5 \text{ mm} \times 2.5 \text{ mm} \times 7 \text{ mm}$ \cite{196, 197}. An analysis of how image voxel size (relative to the scale of flow heterogeneity) influences signal attenuation was conducted. To determine the signal generated by an image voxel, sample points (which represent spins) were scattered evenly in the domain. The spacing between sample points was varied until further spatial refinement produced less than 1% change in the generated IVIM parameters. The IVIM parameters were found to converge when the spacing between sample points was 0.05 mm or smaller. Therefore, all simulations were performed with sample points evenly scattered at an interval of 0.05 mm in the $x$-, $y$-, and $z$-directions. If the sample point was located within a fetal vessel, $\psi$ for the point was sampled from the magnetisation field obtained for the fetal circulation. If the sample point was within the boundary between fetal vessels and villous tree, $\psi$ for that point was sampled from the magnetisation field for a field with a uniform diffusion coefficient of water. As for sample points outside the boundary of the villous tree, $\psi$ for those points were sampled from the magnetisation field for the maternal circulation. Sampling of $\psi$ was performed using the linear interpolation method.

Assuming that each sample point represents a spin particle $j$, the total signal ($S$) from the voxel was derived using the discrete form of Equation 4.14, by summing the magnitude of $\psi$ for each sample point in the voxel:

$$S = \text{Re} \left( \sum_j \psi_j \right)$$

$$= \text{Re} \left( \sum_j A_j e^{i\varphi_j} \right)$$

$$= \sum_j A_j \cos \varphi_j \quad (4.23)$$
Fitting of signal attenuation curve

Magnetisation in the placental subunit was simulated over a range of $G$ (and so $b$) values and the signal attenuation at each gradient amplitude was plotted as a function of $b$. Signal attenuation was obtained by normalising the signal from each $G$ against the signal obtained when $G$ is equal to 0 s/mm$^2$ ($S(0)$). The signal attenuation curve was fitted to a biexponential decay function as described by Equation 4.17 to determine the IVIM parameters $f$, $D$, and $D^*$. Details on how the signal attenuation curve was fitted to a biexponential decay function is provided in Appendix B.3.

4.3.5 Model parameterisation

Clinical studies have used IVIM to visualise blood flow in placentas at approximately 20 weeks [196, 197]. Therefore, we parameterised the model to a 20 week placenta for analysis. Estimates of post-partum thickness and volume of placentas [12] vary between in utero ultrasound measurements [209, 210]. For all Models 1–5, we used post-partum thickness and volume as villous volume and fetal capillary volume have been measured from post-partum placentas using stereological methods [42, 205]. As the parenchymal volume (which consists of villi, villous vessels and IVS) constitutes approximately 80% of the placental volume [42], $\tau$ was estimated by reducing the post-partum placental thickness for a week 20 placenta [12] by 20%. $\epsilon$ was estimated based on a nominal volume of the placenta ($8.67 \times 10^4$ mm$^3$), close to the average post-partum placental volume of $9 \times 10^4$ mm$^3$ [42]. The nominal values and literature range for the domain geometry are listed in Table 4.1.

As week 20 placentas are largely composed of stem villi, and thick and bulbous immature intermediate villi in the absence of long slender terminal branches and terminal convolutes [12, 211], we assumed that the villous trees branch up to an average of 15 generations of villi without any TBs. To obtain a villous tree with approximately 32000 ($\sim 2^{15}$) terminals, the same number of seedpoints ($n_{\text{seed}}$) were implemented in the 3D domain. The villous trunk connecting the chorionic plate and the villous tree has a diameter ($d_s$) of 0.2–1.0 mm [39, 204], and an artery with a diameter ($d_A$) of 0.2–0.4 mm [204]. We assumed that the villous trunk reaches its full length after week 16 and $l_s = 2$ mm at week 20, since $l_s$ measured in a week 16 placenta [39] is the same length estimated for a term placenta [63]. Given $n_{\text{seed}}$, $d_s$, $d_A$, and $l_s$ estimates, the remaining input parameters
(\(d_d/d_p, D_{SR}, l_{min}, \theta_{max}\) and \(f_{dist}\)) were fitted to generate a 3D branching structure with a total villous tree volume between 496–550 mm\(^3\) (0.496–0.550 ml) and a fetal capillary volume between 41.3–45.0 mm\(^3\) (0.0413–0.045 ml) as estimated for week 20 placentas using stereological techniques [42, 205]. The parameters were also fitted to ensure that the Strahler branching ratio lies between 2.19 to 2.83 as determined by Kosanke et al. [43] using reconstructed trees obtained from different stages of gestation.

For Model 1, we chose a network of 400 vessel segments, in which each vessel was assigned a nominal radius of 0.2 mm and a length of 1 mm. A nominal flow velocity of 70.7 mm/s, reflective of the inlet velocity for a week 20 tree [212, 213] was selected.

As a test of concept, a simple 3D tree-like structure with a volume as close as possible to the randomly oriented network of branches was generated for Model 2 using 256 seed points with the method described in Section 3.2.1, with each tree branch having a fixed radius of 0.2 mm. For Model 2, a vessel tree representative of a week 20 placental subunit was also created using the parameters listed in Table 4.1. The key metrics of the generated vessel tree are shown in Table 4.2, and Figure 4.5 shows a representation of the vessel tree in Model 2.

Fetal blood flow into the inlet of the villous vessels (\(Q_{in}^f\)) was fixed at 5 mm\(^3\)/s (0.3 ml/min) based on the estimated blood flow in the umbilical vein [212, 213]. Fetal blood flow was only simulated in the arterial tree, with an outlet pressure prescribed at all terminal ends of the fetal vascular tree (\(P_t\)). As the blood vessels were assumed to be rigid tubes, the value of outlet pressure will only affect the inlet pressure without having much effect on the flow distribution due to flow conservation. Here, we used an outlet pressure of 4000 Pa (30 mmHg) to mimic the pressure estimated for the capillaries in the terminal villi [168].

For Model 4, a hydraulic conductivity field for IVS flow was derived based on the structural properties of a week 20 villous tree generated using parameters listed in Table 4.1. To reflect a representative villous tissue density of 0.6 [42], the size of elements in the sampling grid was estimated as 0.85 mm × 0.80 mm × 0.85 mm. The hydraulic conductivity from the spatially varying field was averaged to derive a representative
Table 4.1: Model parameters for a week 20 placenta. The table includes chosen nominal value and literature range.

<table>
<thead>
<tr>
<th>Symbol</th>
<th>Description</th>
<th>Nominal value</th>
<th>Literature range</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>τ</em></td>
<td>Domain thickness</td>
<td>12 mm</td>
<td>15–22.4 mm</td>
<td>[12, 209, 210]</td>
</tr>
<tr>
<td><em>ε</em></td>
<td>Domain width</td>
<td>8.5 mm</td>
<td>8.5–10.5 mm</td>
<td>[42, 181]</td>
</tr>
<tr>
<td><em>x</em>&lt;sub&gt;v&lt;/sub&gt;</td>
<td>Distance between SA and DV</td>
<td>4 mm</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td><em>d</em></td>
<td>Diameter of SA and DVs</td>
<td>0.5 mm</td>
<td>0.2–0.5 mm</td>
<td>[185]</td>
</tr>
<tr>
<td><em>n</em>&lt;sub&gt;seed&lt;/sub&gt;</td>
<td>Number of seedpoints</td>
<td>32000</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td><em>l</em></td>
<td>Stem length</td>
<td>2 mm</td>
<td>2 mm</td>
<td>[39, 63]</td>
</tr>
<tr>
<td><em>d</em>&lt;sub&gt;s&lt;/sub&gt;</td>
<td>Stem diameter</td>
<td>1 mm</td>
<td>0.2–1.0 mm</td>
<td>[39, 204]</td>
</tr>
<tr>
<td><em>l</em>&lt;sub&gt;min&lt;/sub&gt;</td>
<td>Minimum branch length</td>
<td>0.05 mm</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td><em>θ</em>&lt;sub&gt;max&lt;/sub&gt;</td>
<td>Maximum branch angle</td>
<td>20°</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td><em>f</em>&lt;sub&gt;dist&lt;/sub&gt;</td>
<td>Fractional distance to centre of mass</td>
<td>0.4</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td><em>d</em>&lt;sub&gt;d&lt;/sub&gt;/d&lt;sub&gt;p&lt;/sub&gt;</td>
<td>Daughter to parent branch diameter ratio</td>
<td>0.917</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td><em>d</em>&lt;sub&gt;A&lt;/sub&gt;</td>
<td>Stem villus artery diameter</td>
<td>0.3 mm</td>
<td>0.2–0.4 mm</td>
<td>[204]</td>
</tr>
<tr>
<td><em>D</em>&lt;sub&gt;SR&lt;/sub&gt;</td>
<td>Strahler diameter ratio</td>
<td>1.21</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td><em>D</em>&lt;sub&gt;v&lt;/sub&gt;</td>
<td>Diameter of villous vein</td>
<td>Twice arterial diameter</td>
<td></td>
<td>[206]</td>
</tr>
</tbody>
</table>

Maternal blood flow

<table>
<thead>
<tr>
<th>Symbol</th>
<th>Description</th>
<th>Nominal value</th>
<th>Literature range</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>μ</em></td>
<td>Blood viscosity</td>
<td>4×10&lt;sup&gt;-3&lt;/sup&gt; Pa.s</td>
<td>4×10&lt;sup&gt;-3&lt;/sup&gt; Pa.s</td>
<td>[55]</td>
</tr>
<tr>
<td><em>Q</em>&lt;sub&gt;in&lt;/sub&gt;</td>
<td>Inlet flow</td>
<td>16.3 mm&lt;sup&gt;3&lt;/sup&gt;/s (0.98 ml/min)</td>
<td>16.3 mm&lt;sup&gt;3&lt;/sup&gt;/s (0.98 ml/min)</td>
<td>[213]</td>
</tr>
<tr>
<td><em>κ</em>&lt;sub&gt;empty&lt;/sub&gt;</td>
<td>Maximum hydraulic conductivity</td>
<td>0.52 mm&lt;sup&gt;2&lt;/sup&gt;</td>
<td>∞</td>
<td>–</td>
</tr>
</tbody>
</table>

Fetal blood flow

<table>
<thead>
<tr>
<th>Symbol</th>
<th>Description</th>
<th>Nominal value</th>
<th>Literature range</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Q</em>&lt;sub&gt;f&lt;/sub&gt;</td>
<td>Blood flow into villus artery inlet</td>
<td>5 mm&lt;sup&gt;3&lt;/sup&gt;/s</td>
<td>5.00–5.83 mm&lt;sup&gt;3&lt;/sup&gt;/s</td>
<td>[212, 213]</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(0.30 ml/min)</td>
<td>(0.30–0.35 ml/min)</td>
<td></td>
</tr>
<tr>
<td><em>P</em>&lt;sub&gt;f&lt;/sub&gt;</td>
<td>Blood pressure at vessel tree terminals</td>
<td>4000 Pa</td>
<td>2666–5333 Pa</td>
<td>[168]</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(30 mmHg)</td>
<td>(20–40 mmHg)</td>
<td></td>
</tr>
</tbody>
</table>
4.3. Methods

Figure 4.5: 3D model representation of a week 20 vessel tree. The domain has a thickness $\tau$ and width $\epsilon$. The representative vessel tree was generated from realistic morphometric parameters and fed by a central SA and drained by two DVs.

Table 4.2: Key metrics of a week 20 villous tree and its arterial tree generated using nominal parameters from Table 4.1.

<table>
<thead>
<tr>
<th>Metric</th>
<th>Generated Value (s.d.)</th>
<th>Literature value when available [References]</th>
</tr>
</thead>
<tbody>
<tr>
<td>Maximum generations</td>
<td>23</td>
<td>7–34 [12, 62–64]</td>
</tr>
<tr>
<td>Mean number of generations</td>
<td>15</td>
<td>15 [63]</td>
</tr>
<tr>
<td>Mean branching angle</td>
<td>48.6° (27.7°)</td>
<td>–</td>
</tr>
<tr>
<td>Major/minor branching angles</td>
<td>0.99</td>
<td>–</td>
</tr>
<tr>
<td>Vessel length to diameter ratio</td>
<td>3.01 (0.97)</td>
<td>–</td>
</tr>
<tr>
<td>Vessel diameter/diameter of parent</td>
<td>0.99 (0.06)</td>
<td>–</td>
</tr>
<tr>
<td>Strahler branching ratio</td>
<td>2.39</td>
<td>2.19–2.83 [43]</td>
</tr>
<tr>
<td>Diameter of terminal vessel branches</td>
<td>0.03 mm</td>
<td>–</td>
</tr>
<tr>
<td>Volume of villous tree</td>
<td>547 mm$^3$</td>
<td>496–550 mm$^3$ [42, 205]</td>
</tr>
<tr>
<td>Volume of capillary tree</td>
<td>43.7 mm$^3$</td>
<td>41.3–45.0 mm$^3$ [42, 205]</td>
</tr>
</tbody>
</table>
Table 4.3: Parameters for generating signals from PGSE sequence.

<table>
<thead>
<tr>
<th>Symbol</th>
<th>Description</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>$\gamma$</td>
<td>Gyromagnetic ratio of hydrogen</td>
<td>$2.68 \times 10^8 \text{ rads}^{-1} \text{T}^{-1}$ (42.58 MHzT$^{-1}$)</td>
</tr>
<tr>
<td>$\delta$</td>
<td>Duration of gradient pulses</td>
<td>0.0184 s</td>
</tr>
<tr>
<td>$\Delta$</td>
<td>Time between first and second gradient pulse</td>
<td>0.060 s</td>
</tr>
<tr>
<td>G</td>
<td>Magnitude of gradient pulse</td>
<td>0, $2.46 \times 10^{-6}$, $5.50 \times 10^{-6}$, $9.53 \times 10^{-6}$, $2.13 \times 10^{-5}$, $3.77 \times 10^{-5}$, $4.92 \times 10^{-5}$, $5.89 \times 10^{-5}$, $7.89 \times 10^{-5}$, $8.63 \times 10^{-5}$, $1.02 \times 10^{-4}$, $1.21 \times 10^{-4}$ Tmm$^{-1}$</td>
</tr>
<tr>
<td>$b$</td>
<td>Resultant diffusion sensitivity parameter</td>
<td>0, 0.2, 1, 3, 15, 47, 80, 115, 206, 246, 344, 484 s/mm$^2$</td>
</tr>
<tr>
<td>$t_1$</td>
<td>Time of application of first gradient pulse</td>
<td>0.0208 s</td>
</tr>
<tr>
<td>$TE$</td>
<td>Echo time</td>
<td>0.120 s</td>
</tr>
</tbody>
</table>

uniform hydraulic conductivity of $4.83 \times 10^{-4}$ mm$^2$ for Model 3. As the villous tree is asymmetric, a velocity boundary condition matching a maternal blood flow of $16.3$ mm$^3$/s (0.98 ml/min) into an SA ($Q_{in}$) was applied. As direct flow measurement through the SA is unavailable, we derived $Q_{in}$ from an even distribution of the volumetric flow in the uterine artery [213] into 100 SAs. Following Models 2 and 4, Model 5 adopted the same parameters in Table 4.1 for a week 20 placental subunit. Adopting the same PGSE sequence protocol employed in the clinical studies [196, 197], all simulations were performed with parameters listed in Table 4.3.

4.4 Results

Model solutions for magnetisation in a simple 1D structure agree with analytical solutions that have been previously published in the case of diffusion only, uniform flow only, and a combination of diffusion and uniform flow [198, 202] (Appendix B.2.1). While analytical solutions are applicable for 1D spatially distributed models (representing flow through networks of blood vessels) and 3D models (representing maternal blood flow), when there is a combination of the two models, numerical solutions are necessary and are presented below.

4.4.1 Model 1—Fetal blood flow in randomly oriented vessels

Most IVIM studies assume quasi-random flow in a close to random network of capillaries. The assumption of a quasi-random flow is potentially influenced by the density of the capillary network, the size of the vessels, the rate of blood flow through the network, and
the distribution of the blood vessels being imaged, with \( D^* \) suggested as \(< l > < v > /6\), where \(< l >\) is the mean vessel length of the capillary network, and \(< v >\) is the average flow velocity [195]. Here, we examine these assumptions using Model 1 (Figure 4.4).

Figure 4.6 shows how percentage error in model predictions of \( f \), \( D \), and \( D^* \) vary with the geometric properties of the vessel network. Percentage error (\% Error) was calculated using

\[
\text{% Error} = \left| \frac{\text{Model predicted value} - \text{Theoretical value}}{\text{Theoretical value}} \right| \times 100\%,
\]

where the theoretical value refers to the vessel volume fraction in the voxel for \( f \), diffusion coefficient of water in tissue for \( D \), and \(< l > < v > /6\) for \( D^* \), respectively.

Figure 4.6a shows that the model is increasingly able to predict \( f \), \( D \), and \( D^* \) as the volume fraction of vessels increases with the number of vessels. However, the model is unable to accurately predict \( f \) and \( D^* \) when the number of vessels is less than 10 (vessel volume fraction in voxel \(< 1.45 \times 10^{-3}\)). Similarly, model predictions for \( f \), \( D \), and \( D^* \) improve with the increase in volume fraction of the capillary network resulting from changes in vessel radius and length (Figure 4.6b and c). The model also predicts that \( D^* \) is independent of vessel radius but varies with the mean length of the vessels, which is consistent with the study of Le Bihan et al. [195].

With the increase in flow velocity, percentage error in \( f \) and \( D \) remain low (Figure 4.7a), while percentage error in \( D^* \) generally decreases. Figure 4.7b shows that introduction of diffusion \((D = 2.5 \times 10^{-3} \text{ mm}^2/\text{s})\) to the flow in the vessel network (marked in red) does not appear to have significant effects on parameter estimation for \( f \), \( D \), and \( D^* \) when the flow velocity is above 2.5 mm/s. However, when the flow velocity is below 2.5 mm/s, introduction of diffusion increases the error in \( f \) predictions. The model predictions for \( D \) and \( D^* \) approach the same order of magnitude at such low velocities (Figure 4.7c), rendering IVIM incapable of distinguishing the pseudo-diffusion occurring due to perfusion from molecular diffusion. In these cases (with or without the inclusion of diffusion in the vessel network), the model predicts \( f \) to be close to 0, resulting in a high percentage error for \( f \) under such low flow velocities.
Figure 4.6: The effects of vessel properties of a randomly oriented vessel network on IVIM parameters. Note that the error in $D^*$ is related to an unknown value ($<l><v>/6$). The model is unable to accurately predict $f$ and $D^*$ when the number of vessel is less than 10 (vessel volume fraction in voxel $< 1.45 \times 10^{-3}$) but the percentage error in $f$, $D$, and $D^*$ decrease with the increase in vessel volume, as shown with the respective increase in (a) number of vessels, (b) vessel radius, and (c) vessel length.

4.4.2 Model 2–Fetal blood flow in villous vessel tree structure

Simple 3D vessel tree

Here we investigate under what conditions does flow in a tree-like structure behave like random walk, and whether the signal generated by flow in a tree is affected by its structural and flow properties (Figure 4.4, Model 2). First, a flow velocity of 70.7 mm/s, (as implemented in Model 1) was applied in each branch of a simple 3D tree structure with a volume fraction as close as possible to the randomly oriented vessel network in Model 1. Figure 4.8a shows that the model correctly predicts $D$ (0.84% error) with a slight over-estimation of $f$ (9% error). However, $D^*$ is approximately 99.8 mm$^2$/s, with
4.4. Results

(a) Errors in $f$ and $D$ are low when flow velocity is varied. Error in $D^*$ decreases as flow velocity is increased. (b) Introduction of diffusion to flow in the vessels (marked in red) does not influence model predictions of $f$, $D$, and $D^*$ when the flow velocity is higher than 2.5 mm/s (dotted vertical line). Below 2.5 mm/s, error in $f$ begins to increase with the introduction of diffusion. (c) For flow velocity below 2.5 mm/s, model predictions for $D$ and $D^*$ approach the same order of magnitude, resulting in inaccurate model predictions for $f$.

As vessel diameter varies between branches in a villous vessel tree, we investigated whether vessel diameter in the tree structure affects parameter estimations. All properties of the simple tree were kept constant except for the diameter of vessel branches, whereby the diameter of the stem vessel was changed to 0.3 mm and a Strahler diameter ratio of
Chapter 4. Interpreting magnetic resonance images of placental blood flow

Figure 4.8: The effects of flow in a tree-like structure on IVIM parameters. The tree-like structure has a volume fraction close to the randomly oriented vessel network in Model 1. Note that the error in $D^*$ is related to an unknown value ($\langle l \rangle < v >/6$) and percentage error is plotted using a logarithm scale here. (a) Predictions for $f$ and $D$ remain accurate ($<10\%$ error) for a tree-like vessel network. However, error in predicted $D^*$ is high. (b) When the vessel diameter is decreased with each Strahler order, the model predictions for $f$ and $D$ improve while error in $D^*$ remains high.

1.21 was introduced to determine the vessel diameter in subsequent tree generations. As illustrated in Figure 4.8b, model predictions for $f$ and $D$ have lower percentage errors when the vessel diameters are varied, while $D^*$ remains at a similar order (90 mm$^2$/s) to the tree with uniform vessel diameter (99.8 mm$^2$/s).

A varying velocity field was introduced in the simple tree with varying branch diameters using the method described in Section 4.3.2. Figure 4.9a shows that the percentage error in $f$ is between 20–30%, which is higher compared to the percentage error in $f$ (1.65%) predicted for the same tree with a uniform velocity field. This is because when a varying velocity field was implemented, the speed of flow at the branches in distal branching generations decreases below the threshold velocity value for detection. Reduction in the error of $f$ prediction with the increase in inlet velocity also supports that flow in a branch has to be above a threshold velocity for detection by the IVIM method. Estimates of $D$ remain accurate at all inlet velocities while error in $D^*$ generally decreases with the increase in inlet velocity. Consistent with Model 1, magnitude of $D^*$ increases with the increase in inlet velocity (Figure 4.9b).
4.4. Results

Figure 4.9: The effects of flow velocity in a simple tree on IVIM parameters. Note that the error in $D^*$ is related to an unknown value ($<l>\langle v \rangle/6$). (a) Errors in $f$ and $D^*$ decrease with the increase in inlet velocity, while error in $D$ remains low at all velocities. (b) Like in Model 1, $D^*$ increases with flow velocity.

Week 20 villous vessel tree

Figure 4.10a illustrates how percentage error in parameter estimates vary when a week 20 villous vessel tree was pruned to different branching generations. Estimations of $D$ remain accurate for all branching generations. Like Model 1, the model is unable to accurately predict $f$ for vessel trees with less than 8 branching generations (vessel volume fraction in voxel $< 1.54 \times 10^{-3}$) due to the small volume fraction occupied by the vessel tree. As the vessel tree volume increases with the number of branching generations, accuracy of $f$ estimates improve to approximately 20% error, which is the same order of error demonstrated by the simple 3D tree model (Figure 4.9a). However, the error in $f$ increases again for tree with 14 or more branching generations. The absolute perfusion volume detected from vessel trees with more than 14 branching generations is the same as the perfusion volume estimated for a vessel tree with 14 generations, which suggests that flow in branches beyond 14 generations is too slow to be detected and estimation of $f$ is limited by a flow velocity threshold. The high percentage error in $D^*$ for all branching generations suggests that $D^*$ is not governed by $<l>\langle v \rangle/6$. Except for trees with less than 8 branching generations, which were not considered due to the inaccurate $f$ prediction, $D^*$ generally decreases with the decrease in average flow velocity in the vessel tree as the number of branching generations increases.
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Figure 4.10: The effects of number of branching generations of a week 20 vessel tree on IVIM parameters. Note that the error in $D^*$ is related to an unknown value ($< l > < v > / 6$). (a) Vessel trees with less than 8 branching generations are too small for detection, generating high error in $f$ in these vessel trees. Error in $f$ for vessel trees with 8–12 branching generations are lower at approximately 20% but increases for trees with 14 or more branching generations as the flow in the distal vessel branches are too slow for detection. While $D$ estimates are accurate for all branching generations, $D^*$ estimates remain high for all cases. (b) Except for the vessel tree with less than 8 branching generations, $D^*$ estimate for other trees decrease with the drop in average flow velocity in the vessel as the number of branching generations increases.

4.4.3 Model 3–Maternal blood flow in a uniform porous medium

Maternal blood flow in the placenta could be considered quasi-diffusive in regions of the IVS that contain a dense villous structure. However, there are also regions of the placenta in which flow is likely ‘coherent’, particularly near the SA mouth where a central cavity would allow flow to be largely unobstructed (i.e. the ‘jets’ observed in ultrasound). To investigate how signal attenuation is affected by maternal blood flow characteristics in different regions of the placenta, we assume imaging voxels are completely filled with maternal blood as shown in Model 3 of Figure 4.4.

Voxels downstream of the SA

First we considered regions near the SA mouth, where flow is likely best represented by a uniform porous medium. As shown in Figure 4.11a, a 0.5 mm $\times$ 0.5 mm $\times$ 0.5 mm voxel with its midpoint at $x = 4.25$ mm, $y = 11.25$ mm, and $z = 4.25$ mm was moved along the $y$-direction away from the SA to examine how perfusion in these voxels affect parameter estimations for $f$, $D$, and $D^*$. As the voxel immediately beneath the SA inlet was constrained by the boundary condition implemented on the SA, we only considered...
4.4. Results

Figure 4.11: Schematic representation of voxels downstream of the SA and their flow velocity vectors. (a) 0.5 mm × 0.5 mm × 0.5 mm voxels downstream of the SA are shaded in green. (b) Flow velocity vector at the midpoint of each voxel (red arrows) and streamlines (black lines) are shown on the 2D cross-sectional area in the x-y plane. Flow velocity vectors generally point down the y-direction and decrease in magnitude as the voxel traverses down the shaded region away from the SA.

Figure 4.12: The effects of blood flow in voxels downstream of the SA on IVIM parameters. (a) Parameter estimation of $f$ is either 1 or 0, suggesting that the flow can be represented by a pseudo-diffusion coefficient $D_p$. (b) $D_p$ decreases with the average flow velocity in the voxel and converges to $2.5 \times 10^{-3}$ mm$^2$/s when the average flow velocity drops below 0.5 mm/s.
downstream voxels that were not part of the SA. The voxel size is smaller than the typical voxel size used in IVIM imaging but observations derived from these smaller voxels are translatable to bigger voxels, as long as the nature of flow in the voxels remains the same. Flow velocity in the voxel is considered ‘coherent’ as it points downward in the $y$-axis, but the magnitude decreases as the voxel is moved away from the SA (Figure 4.11b).

Biexponential fitting of the signal attenuation curves generated by these voxels produce an $f$ that is either 1 or 0 (Figure 4.12a), which implies that the signal is attenuated exponentially and is represented by a single pseudo-diffusion coefficient $D_p$. As the voxel is moved downstream of the SA, Figure 4.12b shows that $D_p$ decreases with the average flow velocity in the voxel. When flow velocity drops below 0.5 mm/s, the predicted $D_p$ converges to the diffusion coefficient that was set in the model ($2.50 \times 10^{-3}$ mm$^2$/s), implying that diffusion is the dominant mode of transport in these voxels.

**Voxels spanning between the SA and DV**

A 0.5 mm $\times$ 0.5 mm $\times$ 0.5 mm voxel with its midpoint at $x = 4.25$ mm, $y = 11.25$ mm, and $z = 4.25$ mm was moved along the $x$-axis between the SA and DV, so as to examine the nature of signal generated by flow with varying velocity magnitudes and directions within a voxel scale (Figure 4.13). Figure 4.14a describes how model prediction for $f$, $D$, and $D^*$ vary as the voxel is moved along the $x$-direction away from the SA. Except at the starting position, signal from voxels that are less than 2 mm away from the SA do not fit a biexponential function, and the generated $f$ estimates are outside the range of 0 to 1. Figure 4.14b shows an example of the signal attenuation curve generated by a voxel whose midpoint is 1 mm away from the midpoint of the SA. The signal first decreases and increases again as $b$ is increased. It is not evident how the signal attenuation curve from these voxels are affected by the changes in magnitude and direction of flow velocity within the voxel (Figure 4.13b). However, when the voxel is more than 2 mm away from the SA, the signal tends towards a biexponential fit with $f$ close to 0. Unlike the voxels downstream of the SA (unidirectional flow with varying velocity magnitude), the predicted $D_p$ from these voxels do not correspond with the average flow velocity of the voxels (Figure 4.14c).

**Voxels with increasing size**

Starting from a 0.5 mm $\times$ 0.5 mm $\times$ 0.5 mm voxel with its midpoint at $x = 4.25$ mm, $y = 11.25$ mm, and $z = 4.25$ mm, the voxel size was varied by concurrently increasing
4.4. Results

Figure 4.13: Schematic representation of voxels spanning between the SA and DV and their flow velocity vectors. (a) 0.5 mm $\times$ 0.5 mm $\times$ 0.5 mm voxels spanning the downstream region between the SA and DV are shaded in green. (b) Flow velocity vector at the midpoint of each voxel (red arrows) and streamlines (black lines) are shown on the 2D cross-sectional area in the $x$-$y$ plane. Flow velocity vectors display varying magnitudes and directions as the voxel moves from the SA to DV along the shaded region.

the voxel length in all three directions to investigate how voxel size relative to flow heterogeneity influences signal attenuation (Figure 4.15). As shown before in Figures 4.12a and 4.14a, ‘coherent’ flow in the 0.5 mm $\times$ 0.5 mm $\times$ 0.5 mm voxel gives rise to signals which decay exponentially ($f = 1$). With the introduction of flow heterogeneity with an expansion of voxel size, the generated signal switches to a biexponential decay ($0 < f < 1$) (Figure 4.16a). In these voxels, model predictions for $D$ and $D^*$ vary with the
Figure 4.14: The effects of blood flow in voxels between the SA and DV on IVIM parameters. (a) Estimates for $f$ for voxels that are less than 2 mm away from the SA are outside the range of 0–1 as the signal cannot be fitted with a biexponential function. Beyond 2 mm from the SA, the voxel signal is biexponential in nature and $f$ is close to 0. (b) Signal attenuation curve generated from a voxel whose midpoint is $x = 1$ mm away from the SA. The signal does not fit a biexponential function as it decreases initially and increases again when $b$ is increased. (c) For voxels that are 2 mm or further from the SA, model prediction of $D_p$ do not correspond to the average flow velocity of the voxel.
4.4. Results

Figure 4.15: Schematic representation of voxels with increasing size and their flow velocity vectors. (a) A 0.5 mm × 0.5 mm × 0.5 mm voxel (green) was expanded incrementally to a 7 mm × 7 mm × 7 mm voxel (magenta). (b) Flow velocity vectors (red arrows) and streamlines (black lines) are shown on the 2D cross-sectional area in the x-y plane. Flow velocity vectors display more heterogeneity in magnitudes and directions with the increase in voxel size.

Figure 4.16: The effects of blood flow in voxels with different sizes on IVIM parameters. (a) The voxel with a voxel length of 0.5 mm contains ‘coherent’ flow and its signal decays exponentially while signals from bigger voxels follow a biexponential decay with $f$ between 0 and 1 due to the heterogeneity in flow direction and magnitude. (b) Model estimates for $D$ and $D^*$ vary with the average flow velocity in the voxel.
average velocity of the voxel. While ‘incoherence’ is introduced by flow heterogeneity with
the expansion of voxel size (Figure 4.16), there is no increase in $D$ nor $D^*$ estimates.

**Placental imaging voxels in different placental regions**

Here, a $3.5 \text{ mm} \times 2.5 \text{ mm} \times 7 \text{ mm}$ voxel (which is commonly used in IVIM placental imaging [196, 197]) with a midpoint at a reference position of $x = 4.25 \text{ mm}$, $y = 10.25 \text{ mm}$, and $z = 4.25 \text{ mm}$ (green voxel in Figure 4.17) was moved in the $y$-axis along the region shaded in magenta, and in the $x$-axis along the region shaded in yellow (Figure 4.17) to study how signal is affected by blood flow in different regions of the domain. Signal generated by all voxels exhibit biexponential decay with $b$. As the voxel is moved along the $y$-direction, model predictions for $f$, $D$, and $D^*$ decrease with the drop in average flow velocity of the voxel (Figure 4.18a and b). Estimations for $f$ and $D$ converge to 0 and $2.5 \times 10^{-3} \text{ mm}^2/\text{s}$ (red dotted line in Figure 4.18b), respectively, as the average flow velocity falls below 0.13 mm/s when the midpoint of the voxel is more than 1.5 mm away from the reference position. When the voxel is moved along the $x$-direction, the average flow velocity of the voxel and estimation for $f$, $D$, and $D^*$ also decrease (Figure 4.18c and d). The model predicts that $D^*$ estimates are approximately an order of magnitude bigger than $D$, while $D$ estimates are higher than the diffusion coefficient implemented in the model (red dotted line in Figure 4.18d). The same observations were made when the voxel was moved in the $z$-direction (results not shown).

**Figure 4.17:** Schematic representation of voxels in different placental regions. A $3.5 \text{ mm} \times 2.5 \text{ mm} \times 7 \text{ mm}$ voxel with its midpoint at a reference position of $x = 4.25 \text{ mm}$, $y = 10.25 \text{ mm}$, and $z = 4.25 \text{ mm}$ (green) was translated along the magenta region in the $y$-direction and yellow region in the $x$-direction.
Figure 4.18: The effects of blood flow in voxels from different placental regions on IVIM parameters. (a) Average flow velocity decreases with the movement of the voxel along the $y$-axis away from the reference position. Model prediction for $f$ decreases to 0 as the average flow velocity falls below 0.13 mm/s when the voxel is more than 1.5 mm away from the reference position. (b) $D$ converges to the diffusion coefficient of water implemented in the model (red dotted line) with the movement of the voxel away from the reference position. $D^*$ remains higher than $D$ at all voxel positions. (c) Average flow velocity and predicted $f$ decrease as the voxel was moved along the $x$-axis away from the reference position. (d) While both $D$ and $D^*$ predictions decrease as the voxel was moved further away from the reference position along the $x$-axis, $D^*$ remains higher than $D$. Predicted $D$ is higher than the diffusion coefficient of water set in the model (red dotted line).
4.4.4 Model 4–Maternal blood flow in a porous medium with hydraulic conductivity as function of tree structure

To determine how flow heterogeneity introduced by the structure of the villous tree influences signal generation, model predictions for $f$, $D$, and $D^*$ generated using a porous medium with a hydraulic conductivity field as a function of the tree structure (Figure 4.4, Model 4) was compared to a uniform porous medium (Figure 4.4, Model 3). Like in the uniform porous medium model, a 3.5 mm × 2.5 mm × 7 mm voxel was moved from a reference position of $x = 4.25$ mm, $y = 10.25$ mm, and $z = 4.25$ mm along the yellow region shown in Figure 4.17. As the flow streamlines obtained from the uniform porous medium model (Figure 4.19a) and non-uniform porous medium model (Figure 4.19b) exhibit similar patterns, estimations of $f$, $D$, and $D^*$ vary in the same manner for both models. On a voxel-by-voxel basis, the average conductivity for Model 4 is close to or higher than Model 3, and hence, the average flow velocity and $f$ estimated from Model 4 (marked in red in Figure 4.19c) are consistently higher than Model 3 (in black). Figure 4.19d shows that $D$ and $D^*$ each exhibits small variations between both models.

4.4.5 Model 5–Fetal and maternal blood flow in full placental blood flow model

Using Model 5 (Figure 4.4), we examined how the signal generated by a 3.5 mm × 2.5 mm × 7 mm voxel at the reference position (green voxel in Figure 4.17) affects IVIM parameter estimation when the voxel contains (1) only maternal circulation (no villous tree), (2) maternal blood circulation around villous tree branches without fetal perfusion, and (3) maternal blood circulation plus fetal blood circulation in vessels embedded in the villous tree. When the voxel is fully filled with maternal circulation, the model predicts a biexponential signal decay with an $f$ of 0.23 instead of 1. Introduction of a villous tree structure produces an even lower $f$ of 0.18, as molecular diffusion in the villous tree tissues contributes to the ‘slower’ pseudo-diffusion compartment. The estimated $f$ is also lower than the actual perfusion volume fraction of 0.68. With the introduction of fetal perfusion in vessels embedded within the villous tree structure, the estimated perfusion volume fraction increases by approximately 3.52% ($f = 0.19$) but it is still not reflective of the actual perfusion volume fraction in the voxel. Estimates for $D$ and $D^*$ both decrease when a villous tree without fetal
4.4. Results

Figure 4.19: The effects of blood flow in a non-uniform porous medium on IVIM parameters. Flow streamlines generated by the (a) uniform porous medium model (Model 3), and (b) non-uniform porous medium (Model 4) follow similar pattern. (c) Model predictions for Model 3 are in black while results for Model 4 are in red. While average flow velocity and predicted $f$ follow the same trend for both models, the parameters are higher in Model 4. (d) Model predictions for Model 3 are in black while results for Model 4 are in red. Predictions for $D$ and $D^*$ generally exhibit the same trend with small variations between the two cases.

When all flow components were included in Model 5, Figure 4.21 shows that the model predicts a lower $f$ and $D$, and a higher $D^*$ for a voxel located near the chorionic plate (midpoint of voxel at $x = 4.25$ mm, $y = 2.25$ mm, and $z = 4.25$ mm) as compared to a voxel near the basal plate (at reference position).
Chapter 4. Interpreting magnetic resonance images of placental blood flow

Figure 4.20: The effects of placental blood flow components on IVIM parameters. A 3.5 mm × 2.5 mm × 7 mm voxel with midpoint at $x = 4.25$ mm, $y = 10.25$ mm, and $z = 4.25$ mm was used for analysis. The model predicts a decrease in $f$ with the introduction of a villous tree to the voxel containing only maternal circulation, and an increase of 3.52% in $f$ when fetal perfusion was introduced. The estimated $f$ for all cases underestimate the actual perfusion volume fraction in the voxel ($f = 0.68$ as indicated by red dashed line). Both $D$ and $D^*$ decrease with the introduction of villous tree structure and fetal perfusion. Estimates for $D$ in all three cases are above the diffusion coefficient of water set in the model (blue dotted line).

Figure 4.21: The effects of voxel location on IVIM parameters. (a) Voxel near the basal plate is shaded in green (reference position) while voxel near the chorionic plate is shaded in magenta. (b) Apart from $D^*$, $f$ and $D$ generated by the voxel near the chorionic plate show percentage drop when compared to the voxel near the basal plate.
4.5 Discussion

We have presented a model of PGSE signal generation in a placental subunit, which captures how signals are influenced by the geometry and flow properties within the placenta. While the principles of the IVIM technique are well understood and applied to perfusion imaging in different organs such as the brain [214] and liver [215], where blood capillaries embedded in stationary tissues display pseudo-diffusive behaviour, it remains unclear whether the classical assumptions of IVIM are applicable for imaging perfusion in the independent maternal and fetal blood circulatory systems of the placenta. Using the current model, we explored how properties of each circulatory system contribute to PGSE signal generation on an individual as well as a combined basis. This provides an approach to explore the physiological significance of IVIM parameters estimated from placental images, and evaluate the applicability and limitations of IVIM technique for perfusion imaging of the placenta.

Fetal blood flow

Our model predictions compare well to previously published studies on perfusion imaging of micro-circulatory networks in stationary tissues [195]. By applying the IVIM technique in Model 1, which is a randomly oriented vessel network embedded within a stationary domain undergoing molecular diffusion, the model is able to accurately capture the volume of vessels as the perfusion fraction ($f$), and the diffusion coefficient of water implemented in the model as the diffusion coefficient in stationary tissue ($D$). Consistent with the findings published by Le Bihan et al. [195], the apparent diffusion coefficient of moving blood ($D^*$) predicted in Model 1 is related to the mean vessel length and the mean flow velocity in the vessels (Figures 4.6 and 4.7).

In Model 2, we represented the fetal blood vessels as tree-like structures to render physiological realism to the fetal vessels in the placenta. By comparing a tree-like structure with a volume close to a randomly oriented vessel network, our model shows that fetal blood flow in a tree-like structure also exhibits pseudo-diffusive properties like in a randomly oriented network of capillaries. The model is able to adequately predict the perfusion fraction in the tree-like architecture (approximately 10% error) (Figure 4.8a) when the same uniform flow velocity was implemented in both models. The percentage error in predictions for $f$, and $D$ are low even when the diameter of the vessels were varied in both Models 1 (Figure 4.6b) and 2 (Figure 4.8b). Consistent
with the findings from Le Bihan et al. [195], \(D^*\) is independent of the diameter of the vessels. However, IVIM parameters are affected by the density of the vessels. As shown in Model 1, accuracy of model predictions for \(f\), \(D\), and \(D^*\) all improve with the increase in vessel volume (Figure 4.6). However, when the density of vessels occupies a voxel fraction of less than \(1.54 \times 10^{-3}\), such as when there are less than 10 vessels in Model 1 (Figure 4.6a), or when the week 20 villous tree is pruned to below 8 branching generations in Model 2 (Figure 4.10a), the small volume fraction of the vessels becomes difficult to detect and would likely require a higher sampling resolution for reliable prediction of \(f\).

This is consistent with the observations made by Pekar et al. [216], who used computer simulations to demonstrate the difficulty in estimating small perfusion fractions from a set of gradient-sensitised images. With high prediction errors for \(f\) at such small vessel densities, the associated \(D\) and \(D^*\) estimates are meaningless.

Apart from vessel geometry, the accuracy of model predictions is also affected by the velocity of blood flow in the vessels. When the flow velocity in the blood vessels is high, the contribution to signal attenuation due to their pseudo-diffusive behaviour is more distinguishable from the molecular diffusion in stationary tissue, thereby increasing accuracy in model predictions for \(f\) and \(D\) (Figure 4.7a and Figure 4.9). However, when flow velocity decreases below \(2.5\) mm/s, the resultant \(D^*\) drops to the same order of magnitude as molecular diffusion (\(D\)) and the IVIM technique is unable to accurately separate blood vessels from tissue (Figure 4.7). This is also evident when the uniform flow velocity in the simple 3D tree model was replaced with a varying flow velocity field generated based on the flow resistance of the tree structure in Model 2, whereby the percentage error in \(f\) is approximately 20–30\% (Figure 4.9) as the distal branches of the tree structure with flow velocity below the threshold velocity cannot be adequately captured by the IVIM technique. Similarly, the IVIM technique can only detect perfusion volume in week 20 vessel trees up to 14 branching generations, and flow in subsequent branching generations is indistinguishable from molecular diffusion in tissue. From a clinical perspective, the accuracy of perfusion fraction quantification is limited by the density of blood vessels and the velocity of flow within the vessels.

When perfusion volume and velocity are above their respective threshold values, the model is able to accurately predict \(D\) but the percentage error in \(D^*\) is high in Model 2. Even though this suggests that \(D^*\) in tree-like structure is not simply governed by \(<l><v>/6\) like the flow in a randomly oriented vessel network, the model predicts
that $D^*$ increases with the average flow velocity in the voxel (Figures 4.7c, 4.9b, and 4.10b). Hence, we expect a distribution plot of $D^*$ estimated for each voxel to provide a spatial map of the relative flow velocity in different regions of the tissue, with higher $D^*$ representing higher average flow velocity and vice-versa.

Maternal blood flow
Simulations assessing signal generation by the maternal circulation in the IVS and analysis of the resultant IVIM parameter estimations were conducted using Model 3. Maternal blood flow enters the IVS in a ‘jet-like’ manner and circulates through the IVS with a range of directions and velocities. It is unclear if maternal blood flow in the IVS exhibits pseudo-diffusive behaviour and whether a level of ‘incoherence’ is required for pseudo-diffusive behaviour to be observed. In voxels downstream of the SA inlet where flow is mostly unidirectional with varying velocity magnitudes, we observed that the generated PGSE signal decays exponentially as a function of $b$ (Figure 4.12a). The decay constant is governed by a pseudo-diffusion coefficient $D_p$ which decreases with the average velocity of flow in the voxel, and converges to the diffusion coefficient implemented in the model when transport becomes dominated by diffusion when the voxel was moved away from the SA inlet (Figure 4.12b). The model suggests that while ‘coherent’ flow on a voxel level exhibits pseudo-diffusive behaviour, the signal attenuation curve generated by such voxel is better fitted with an exponential function. However, if the curve is fitted to a biexponential function as in the case of IVIM, the resultant $f$ estimate has to be considered in association with $D$ and $D^*$ estimates. For instance, when $f$ is estimated as zero, it does not necessarily signify that the voxel is devoid of moving blood. If the $D$ estimate for the voxel is higher than the diffusion coefficient of water in tissue, it would suggest a completely opposite scenario whereby the voxel is completely filled with moving blood. Hence, caution is required when interpreting parameter estimates using the IVIM technique.

When the voxels contain varying flow directions and magnitudes, it was observed that the resulting signal profiles do not follow a consistent decay pattern. In some voxels, a multimodal pattern (e.g. Figure 4.14b), which was previously suggested by Moore et al. [197], was observed while other voxels exhibited a biexponential decay with $f$ close to 0 (Figure 4.14a). For voxels that do not generate a biexponential signal decay, no meaningful interpretation can be made from the estimated $D$ and $D^*$. As for voxels with signals that decay exponentially, the associated diffusion coefficient does not appear as a
simple function of the average flow velocity (Figure 4.14c). It is likely that the variance in flow directions adds ‘incoherence’ to the flow, and contributes to the pseudo-diffusive behaviour of flow in the voxel. Further investigation would be required to assess the relationship between flow heterogeneity and the type of generated signal so as to determine an appropriate function to better quantify the nature of flow in such voxels.

When the voxel size was incrementally increased to include more heterogeneity in flow, we observed that the generated signal starts to consistently display a biexponential decay (Figure 4.16a). Also, when the actual voxel dimension used in PGSE imaging was implemented in the model, the signals generated by these voxels in different regions of the domain also display the same decay pattern (Figure 4.18a and c). In these voxels, we identified that $f$, $D$, and $D^*$ generally decrease with decreasing average flow velocity in the voxel (Figures 4.16, 4.18). In voxels where advection is the dominant mode of transport, $D$ estimates are higher than the diffusion coefficient of water in tissue set in the model but remain lower than the estimated $D^*$. In view of these findings, we propose that the flow in such voxels is ‘compartmentalised’ into a ‘faster’ and a ‘slower’ group based on their apparent pseudo-diffusion coefficients, rather than a ‘diffusion’ and ‘perfusion’ component. The ‘faster’ moving blood (represented by a higher apparent diffusion coefficient $D^*$) gives rise to signal attenuation at low $b$ values, while the ‘slower’ moving blood (represented by a lower apparent diffusion coefficient $D$) gives rise to signal attenuation at high $b$ values. In these cases, $f$ is likely representative of the fraction of ‘faster’ moving blood in the voxel, rather than the perfusion fraction of the voxel. An implication is that the $f$ estimated from the IVIM technique can be misleading, and potentially result in the mis-interpretation of a voxel as containing a perfusion and a diffusion compartment even when it is completely filled with maternal blood. This further emphasises the necessity to take into account the values of $D$ and $D^*$ when considering the estimated $f$ value.

In voxels with low flow velocity where the flow is dominated by diffusion, PGSE signals approach a biexponential fit with $f$ close to 0, and the signal attenuation profiles are identical to the profile generated by voxels with tissues undergoing molecular diffusion. This property is likely to complicate the image analysis process as it is difficult to differentiate regions with low maternal blood flow from tissue regions without any blood flow. For example, the boundary between the chorionic plate and the IVS may not be obvious in the placental IVIM images and most likely will result in an underestimation
of the actual placental boundary on the fetal side. Also, perfusion fraction in regions with densely packed villous tree are also likely to be underestimated since maternal blood flow may be slowed down while percolating through the dense tissue network, rendering it invisible to IVIM imaging.

**Fetal and maternal blood flow in the placenta**

Having explored the different flow components of the placenta on a separate basis, the independent blood flow systems were combined to study the signal attenuation due to blood flow in the placenta using Model 5. With the introduction of a villous tree structure (with molecular diffusion of water in stationary tissue) to a voxel filled with maternal blood, the signal attenuation curve retained its biexponential nature, but generated a smaller $f$ since more of the voxel volume is occupied by the diffusion (or ‘slower’ moving) component (Figure 4.20). When fetal blood flow was incorporated into the villous tree structure to assess its influence on signal generation, additional pseudo-diffusive behaviour arising from the fetal blood flow increases the model prediction for $f$ with small changes in $D$ and $D^*$ (Figure 4.20). Even though the increase in perfusion volume due to fetal blood flow is captured with the increase in $f$ estimate, the model prediction for perfusion fraction is still an underestimate of the actual volume of perfusion in the voxel. Underestimation of $f$ has previously been reported in an *in vivo* IVIM study of placentas ranging from week 20 to term [196]. The study predicted an average $f$ of 0.26 which is lower than the fractional volume of the IVS relative to the total volume of villi and IVS measured using stereological methods [217]. A possible explanation for this underestimation is that the IVIM technique quantifies both fetal and maternal flow into a ‘faster’ and a ‘slower’ pseudo-diffusion component, rather than a diffusion and perfusion compartment. As the $D$ estimates generated by Model 5 are higher than the diffusion coefficient of water in tissue that was prescribed in the model, this suggests that the average speed of flow (fetal or maternal) in the ‘slower’ pseudo-diffusion group is moving faster than molecular diffusion and hence, would encompass a bigger voxel fraction including that occupied by the villous tissue (which is undergoing molecular diffusion). This thereby results in a lower prediction of $f$ as compared to the actual perfusion fraction.

In a study conducted by Moore et al. [196], $D$ was estimated as $1.7 \times 10^{-3}$ mm$^2$/s and $D^*$ as $57 \times 10^{-3}$ mm$^2$/s from IVIM measurements of *in vivo* placentas. Since the IVIM measurements were obtained from averaging the values from a range of placentas of different ages, it is unlikely for the current model, which was parameterised to week 20,
to generate quantitatively comparable predictions. However, our model predicts that $D^*$ is an order of magnitude greater than $D$ as estimated in the previous study [196]. As these IVIM parameters are likely to reflect the volume, velocity and average path length of blood flow in both the fetal and maternal circulation, further investigation will be required to understand how flow is separated into the ‘faster’ and ‘slower’ pseudo-diffusion compartments and how fetal and maternal circulations each contributes to the lumped parameters $f$, $D$, and $D^*$.

The IVIM technique has been applied in some studies to understand the perfusion pattern in normal and compromised placentas [196, 197, 218]. Consistent with previous findings, our model predicted two zones of perfusion in a normal placenta, with the region nearer to the basal plate having a higher perfusion fraction than the region nearer to the chorionic plate. The higher perfusion fraction in the basal region coincides with the entry of fast moving maternal blood into the placenta. As maternal blood circulates through the IVS, it loses speed and may even slow down to diffusion scale near the chorionic plate. This explains why the perfusion fraction predicted for the voxels near the chorionic plate is smaller than for the voxels near the basal plate, and is mainly attributed to the fetal vessel flow volume. With the decrease in flow volume and/or velocity of incoming maternal blood in IUGR placentas [219], the model suggests that the perfusion fraction at the basal region will be lower than normal placentas, while the perfusion fraction in the chorionic region will be higher than the normal placentas with the increase in fetal capillarization in adaptation to the decreased maternal blood flow. A more homogeneous distribution of perfusion fraction across different regions of IUGR placentas is hence expected based on the model’s behaviour, and this coincides with observations made from IVIM images of IUGR placentas where two zones of perfusion are less apparent [197].

**Model limitations**

In this model, maternal blood flow was represented by flow in a porous medium. While we have shown that incorporation of a conductivity field based on the structure of the villous tree (Model 4) provides similar predictions for IVIM parameters as a uniform conductivity field (Model 3), there may be more variance in flow velocities and path-lengths as maternal blood flows between villous trees and through the gaps between the villous tree branches in the placenta, which cannot be adequately captured with a porous medium model. Since the model currently does not include detailed description of how the flow path of maternal blood is obstructed or affected by individual branches
of the villous tree, we have taken a simplified approach of superimposing fetal blood flow and villous tree structures over the maternal blood flow field. Furthermore, we assumed that the variability of diffusion coefficient due to the different pore sizes in the IVS is negligible. Since the diffusion coefficients $D$ and $D^*$ are both related to the average path-length relative to pulse sequence timings, all these factors may affect the estimations of $f$, $D$, and $D^*$. Despite these limitations, the current model is sufficient to demonstrate the general applications of IVIM in different flow regions of the placenta. However, these limitations should be considered when undertaking further extensions of the model to better quantify how IVIM parameters relate to blood flow properties in the placenta.

We have assumed a fixed orientation of the placental subunit with respect to the axis of gradient application. As the location of the placenta in the uterus varies between subjects, it will be insightful to analyse how signal attenuation is affected by the different orientation of the placental subunit to the axis of gradient application and explore the possibility of standardising the orientation of gradient application with respect to the placenta for better image visualisation. Moreover, as MRI is sensitive to motion due to the use of magnetic gradients, it will also be useful to investigate the effects of pulsatile blood flow in the IVS [220] and in the fetal circulation [221] using the model to assess the necessity to include such features in future models.

### 4.6 Summary and conclusions

In this chapter, we used the placental blood flow model to examine the effects of maternal blood flow and fetal blood flow on PGSE signal generation, and assessed the applicability of the IVIM technique for blood flow visualisation in the human placenta. The modelling framework is able to accurately predict perfusion fraction and flow properties for a tissue with a random vessel network in accordance to LeBihan’s IVIM theory [195]. The model revealed that flow in a tree-like structure (like in the fetal circulation) also exhibits pseudo-diffusive behaviour provided that the vessel density and flow velocity are above the detection limit. However, $D^*$ does not follow $< l > < v > / 6$ as proposed for flow in a network of random vessels, even though it increases with flow velocity. Voxels containing coherent flow (e.g. maternal blood near the SA inlet) produces an exponential signal decay governed by a pseudo-diffusion coefficient that decreases with the flow velocity magnitude, while voxels containing maternal blood with varying flow
velocity magnitudes and directions sometimes generate signal with a multimodal or biexponential pattern. Overall, interpretation of $f$ has to be performed with caution in conjunction with $D$ and $D^*$, as the maternal blood flow model (Model 3) and full placental blood flow model (Model 5) both suggest that $f$ estimates represent the voxel fraction of ‘faster’ moving blood, rather than the fraction of perfusion. Although some of these observations were made from separate considerations of the fetal and maternal blood flow system, they are relevant and applicable for interpreting flow nature in different regions of the placenta, and most importantly, can guide clinicians in better understanding what they are seeing in placental images.

In the next chapter, we will couple the maternal blood flow model with an oxygen transport model to investigate the influences of villous tree geometry on oxygen transport and fetal oxygen uptake from the IVS.
Chapter 5

Predicting oxygenation in a placental subunit

5.1 Motivation

Numerous pregnancy complications such as pre-eclampsia and IUGR have been associated with placental malfunction [222]. Morphological abnormalities such as reduced volume and surface area of terminal villi [14, 16], decreased villus length [14, 16], increased trophoblast epithelium thickness [14], and abnormally sparse fetal capillary networks [18, 143], have been identified in pathological placentas. However, it is not fully understood how these villus abnormalities translate to oxygen exchange dysfunction in the placenta. With the difficulties in directly measuring placental function, computational models are useful tools to investigate how observed pathologies in villous tree structure influence placental oxygen transfer.

As discussed in Section 2.7, early models of gaseous exchange within the placenta placed particular emphasis on modelling the exchange of oxygen between the maternal and fetal bloodstreams at terminal villi [50, 51]. However, they neglect the spatial flow characteristics of the maternal circulation and its resultant impact on gaseous exchange. Later models considered maternal flow in the IVS as a porous medium [53, 54]. Erian [53] modelled the intervillous space in 2D, and through the assumption that the villous tree is significantly deformed by maternal flow, flow-dependent tissue permeability was included into their model. Chernyavsky et al. [55] combined a 3D axisymmetric porous
medium approach and first order solute uptake kinetics with the use of a homogenised villous tree (uniform porosity throughout). Serov et al. [56, 57] introduced a stream-tube model of oxygen exchange in a placental subunit, and assumed that maternal blood flow streamlines follow villous branches with oxygen uptake occurring across the villous branch surfaces which extend uniformly along the branch length from the central cavity to the DV. These models have suggested different estimates of optimal villous tissue density for placental efficiency, with estimates from the stream-tube model comparing well to stereological data [56]. However, these models do not explicitly account for the branching architecture of the villous tree, which has been suggested to improve the accuracy of model predictions [171]. A recent multiscale model of the placental vasculature [168] suggested that vascular branching may influence placental efficiency, but oxygen exchange was not modelled in this work. Lecarpentier et al. [170] recently used the Navier-Stokes equation to model blood flow between ‘rigid’ tissue obstacles in a 2D representation of the IVS. Even though this approach includes significant geometrical detail, it is computationally expensive and so does not lend itself to large scale simulations of blood flow and/or nutrient transport in the whole placenta.

Current models of oxygen exchange in the placenta typically assume a uniform oxygen exchange potential at any point within the villous tree structure. For example, there is no distinction between terminal and stem villi in terms of exchange barrier thickness. The harmonic mean barrier thickness between maternal and fetal blood varies through the placenta with the thinnest barriers (and most mature terminal villi) residing peripherally in the placental subunit [130, 223]. It has been theorised that peripheral villi are more effective gas exchangers than central ones, as they reside in a lower oxygen environment than central villi. A central-to-peripheral gradient in oxygen concentration has been confirmed in primate placental subunits [128, 129], which is consistent with hypothesised blood flow distributions [104], and has been visualised in humans using BOLD [131]. To date, computational models of placental oxygen exchange have not been able to account for these structural and functional inhomogeneities.

In Chapter 3, we identified how maternal blood flow in the IVS is influenced by the geometry of the villous tree. In this chapter, we present a computational framework which builds on our maternal blood flow model to simulate oxygen transport in a placental subunit. This oxygen transport model aims to bridge the gap between detailed geometric models [168, 170] and highly smoothed homogeneous models [55–57, 169]. This model
will be used to predict how oxygen uptake in a human placental subunit is influenced by a non-uniform porosity distribution over the villous structure, and whether regional variations in oxygen uptake capacity influence placental efficiency. Like the maternal blood flow model, this modelling framework is presented in 2D and parameterised using morphological data. Work discussed in this chapter has been published [172].

5.2 Methods

Oxygen in the IVS is transported by advection (by maternal blood) and diffusion before it is taken up by the villous trees. The maternal blood flow model described in Chapter 3 was used to predict how oxygen carried by maternal blood is advected in the IVS. The solution procedure to find the distribution of oxygen in the IVS and fetal oxygen uptake rate is shown in Figure 5.1. Each model component is described in turn below.

**Figure 5.1:** Components of oxygen transport model. Maternal blood flow velocity field was generated from the maternal blood flow model to predict how blood is advected in the IVS. Oxygen uptake was distributed over the oxygen transport mesh based on location of villous branches and TBs, and the distribution of oxygen and fetal oxygen uptake were derived by solving the advection-diffusion-reaction equation.

5.2.1 Model geometry

We used the same 2D model geometry described in Chapter 3, which consists of a villous tree, and a maternal circulation made up of a central SA and two DVs. Such geometry is representative of a longitudinal cross-section of a 3D placental subunit.
5.2.2 Modelling oxygen transport

**Governing equation**

Oxygen concentration in the maternal blood in the IVS ($C_m$) was described by the advection-diffusion-reaction equation:

$$\frac{\partial C_m}{\partial t} + \mathbf{V} \cdot \nabla C_m = D_{O_2} \nabla^2 C_m - \alpha (C_m - C_f),$$  \hspace{1cm} (5.1)

where $t$ is time, $\mathbf{V}$ is the maternal blood flow field derived from the maternal blood flow model described in Section 3.2.2 (Figure 5.1, step 1), $D_{O_2}$ is the diffusivity of oxygen in maternal blood, $C_f$ is the oxygen concentration in the fetal bloodstream and $\alpha$ is the oxygen uptake constant. As oxygen was assumed to be taken away immediately by the fetal bloodstream upon uptake, $C_f$ was set as constant.

**Finite element mesh and solution procedure**

Equation 5.1 was solved in step 3 (Figure 5.1) for oxygen distribution in the IVS using the Lagrange-Galerkin finite element method (Appendix C) [208], where for every timestep, the departure point of blood at each mesh node was traced and $C_m$ at that mesh node was initiated with the concentration at the blood’s departure point at the start of the timestep. The updated oxygen concentration field was then used as an initial condition to calculate the new oxygen distribution resulting from diffusion and uptake using the Galerkin finite element method.

A finite element mesh consisting of bilinear elements was used to solve Equation 5.1 (the oxygen transport mesh). The spatial resolution of this mesh was the same as the maternal flow mesh described in Chapter 3. Typical sizes for the timestep used in this model to achieve temporal convergence ranged from $5 \times 10^{-6}$ s to $5 \times 10^{-3}$ s. The model was solved to obtain the steady state.

**Oxygen uptake by terminal villi**

At the terminal villus level, oxygen uptake was assumed to occur over the whole region bound by the TB. Using the endpoint location of each intermediate villus feeding a TB, oxygen uptake constant ($\alpha$) was distributed to each of the nodes of the sampling grid element using a bilinear interpolation function. The resulting $\alpha$ distribution was sampled over the oxygen transport mesh when the model was solved.
5.2. Methods

**Oxygen uptake by stem and intermediate villous branches**

As the stem and intermediate villi were explicitly modelled as discrete branches, \( \alpha \) was assigned at the start and end of every stem and intermediate villous branch. For each villus, \( \alpha \) was distributed between the nodes of the oxygen transport mesh element containing the villus. The \( \alpha \) distribution for TBs and non-terminal villi were summed to obtain an overall \( \alpha \) distribution and used to solve Equation 5.1 (Figure 5.1, step 2).

**Boundary conditions**

The oxygen concentration of the maternal blood entering the domain through the SA was fixed as a constant (\( C_{in} \)). At all other boundaries, a zero diffusive flux was imposed (\(-D_{O_2}\nabla(C_{m})\cdot \mathbf{n} = 0\)), where \( \mathbf{n} \) is the outward facing normal to the boundary), which implies that oxygen was carried across the boundary at the DVs via the advective component of the solution.

**Model outputs**

Key model outputs were the flow and pressure distributions of maternal blood, the rate of oxygen uptake by the villous tree and the distribution of oxygen concentration in the IVS (Figure 5.1). The rate of oxygen uptake by the villous tree was computed as the difference between the concentration fluxes at the inlet SA and the outlet DVs, while the average oxygen content in the IVS was calculated by dividing the sum of oxygen content at all finite element nodes in the domain by the total number of finite element nodes.

5.2.3 Model parameterisation

Following the model geometry and maternal blood flow parameters in Chapter 3, the current model was parameterised to the term placenta as listed in Table 3.2. Nominal parameters related to the oxygen transport model and values reported from the literature are shown in Table 5.1. The diffusivity of oxygen used in the model was based on measurements made from blood [224], which accounted for the differences in oxygen diffusivity in plasma and haemoglobin. An estimate of the oxygen content in the fetal bloodstream (\( C_f \)) was derived from oxygen measurements obtained from the umbilical artery immediately after delivery [225], while the oxygen content in the SA (\( C_{in} \)) was based on the median partial pressure of oxygen (pO\(_2\)) in uterine artery collected from pregnant women during their cesarean section [226]. The conversion of pO\(_2\) to oxygen concentration is detailed in Appendix D. \( \alpha \) is predominantly defined by the diffusing
Table 5.1: Oxygen transport model parameters for a 2D term placental subunit. The table also includes chosen nominal value and literature range for each parameter.

<table>
<thead>
<tr>
<th>Symbol</th>
<th>Description</th>
<th>Nominal value</th>
<th>Literature range</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>$D_{O_2}$</td>
<td>Diffusivity of oxygen in blood</td>
<td>$1.62 \times 10^{-3}$ mm$^2$/s</td>
<td>$1.62 \times 10^{-3}$ mm$^2$/s</td>
<td>[224]</td>
</tr>
<tr>
<td>$C_f$</td>
<td>Oxygen concentration in fetal blood</td>
<td>$3.43$ mol/m$^3$</td>
<td>$3.43$–$8.17$ mol/m$^3$</td>
<td>[225]</td>
</tr>
<tr>
<td></td>
<td>(8.72$\times 10^{-2}$ ml/ml)</td>
<td>(8.72$\times 10^{-2}$–2.08$\times 10^{-1}$ ml/ml)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>$\alpha$</td>
<td>Oxygen uptake constant</td>
<td>$0.38$ s$^{-1}$</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td>$C_{in}$</td>
<td>Oxygen concentration entering through SA</td>
<td>$6.44$ mol/m$^3$</td>
<td>$6.40$–$6.52$ mol/m$^3$</td>
<td>Appendix D, [226]</td>
</tr>
<tr>
<td></td>
<td>(1.64$\times 10^{-1}$ ml/ml)</td>
<td>(1.63$\times 10^{-1}$–1.66$\times 10^{-1}$ ml/ml)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

capacity of the exchange barrier [52, 132, 133], which is proportional to the average surface areas of the villi and fetal capillaries, and inversely proportional to the harmonic mean diffusion thickness of the villous membrane [50, 52, 132, 133]. Although the properties of the exchange barrier vary in different regions of the placenta, an arbitrary constant of 0.38 s$^{-1}$ was set as the nominal case and subsequently varied to examine how regional variation in $\alpha$ affects oxygen uptake efficiency.

5.3 Results

5.3.1 Model verification

To verify the accuracy of the current model, we parameterised our model as a uniformly porous medium with the parameters used in Chernyavsky et al. [55]. Although based upon different methodologies, our model is able to reproduce comparable results with those generated by Chernyavsky et al. [55]. Detailed descriptions of model parameterisation and the generated results are in Appendix E.

5.3.2 Baseline case and consistency with previous studies

Function of a placental subunit

The model makes several predictions on the whole placentome scale that are directly comparable to previous experimental and modelling studies. Table 5.2 shows model predictions of oxygen exchange metrics in the placental subunit under baseline conditions (using nominal parameter values). The predicted oxygen uptake rate is $6$ mm$^3$/s.
Table 5.2: Comparison of predicted oxygen exchange metrics in a placental subunit under baseline conditions with literature data.

<table>
<thead>
<tr>
<th>Metric</th>
<th>Predicted value</th>
<th>Literature range</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Oxygen uptake in subunit</td>
<td>6 mm³/s</td>
<td>3–10.5 mm³/s</td>
<td>[227]</td>
</tr>
<tr>
<td></td>
<td>(0.36 ml/min)</td>
<td>(0.18–0.63 ml/min)</td>
<td></td>
</tr>
<tr>
<td>Average pO₂ in IVS</td>
<td>4400 Pa</td>
<td>2400–4520 Pa</td>
<td>[226, 228, 229]</td>
</tr>
<tr>
<td></td>
<td>(33 mmHg)</td>
<td>(18.0–33.9 mmHg)</td>
<td></td>
</tr>
<tr>
<td>Ratio of pO₂ in venous return to IVS</td>
<td>0.90</td>
<td>1.48</td>
<td>[226]</td>
</tr>
</tbody>
</table>

(0.36 ml/min) per placental subunit, which is close to the mean fetal and placental oxygen consumption of 6.17 mm³/s (0.37 ml/min) per subunit as measured in women undergoing elective cesarean section [227]. The average pO₂ in the IVS predicted by the model is 4400 Pa (33 mmHg), near the higher limit of the range reported in the literature [226, 228, 229]. The ratio of pO₂ in venous return to the IVS is different from the ratio recorded by Schaaps et al. [226] for pO₂ in the uterine vein to the IVS. Their study attributed higher measured pO₂ in the uterine vein to the vascular anastomoses in the myometrium. Since such anastomoses were not incorporated in the current model, the ratio of pO₂ in venous return to IVS is expected to be different and should be less than 1.

Spatial variation in oxygen uptake

Figure 5.2a illustrates the predicted oxygen distribution. Consistent with the ‘physiological concept’ proposed in cine-angiography and injection studies [128], the model predicts a region of high oxygen content where oxygen-rich blood is projected into the IVS from the SA, a decrease in oxygen content due to uptake by villous tissue as maternal blood percolates through tissue, and return of oxygen-depleted blood to the DVs. To assess model predictions against studies hypothesising a central-to-peripheral oxygen gradient [104, 121, 223, 230, 231], we divided the domain into equally sized regions representing central and peripheral tissue, as well as basal, mid-placenta, and chorionic tissue (Figure 5.2a). Figure 5.2b shows higher average oxygen concentrations in the central regions compared to the peripheral regions, and decreasing concentrations as one moves from the basal to chorionic surfaces of the placenta, which is consistent with previous studies.
Chapter 5. Predicting oxygenation in a placental subunit

Figure 5.2: Oxygen distribution predicted for a placental unit under baseline conditions. 
(a) The oxygen concentration field is normalised by oxygen concentration of maternal blood entering the IVS. Oxygen delivered by maternal blood from the SA into the IVS is taken up by villi as maternal blood percolates radially through villous tissue. Maternal blood that is low in oxygen is then drained through the DVs. (b) Comparison of the average partial pressure of oxygen in the six different regions as marked in (a) demonstrates a decreasing central-to-peripheral oxygen concentration gradient as well as a gradient from the basal (uterine) to chorionic sides of the placenta.
5.3. Results

Figure 5.3: The effects of terminal villous tissue density ($\phi_{TV}$) on oxygen uptake. The normal range of oxygen uptake rate determined from the literature is shaded. The model predicts an ‘optimum’ uptake rate for $\phi_{TV} \approx 0.15$, and predicts low oxygen uptake efficiency when surface area for oxygen uptake is small (at low $\phi_{TV}$) or when terminal villi present a high impedance to maternal blood flow (at high $\phi_{TV}$). The dotted line indicates the nominal value of $\phi_{TV}$.

Optimal villus density

Previous studies have assessed the relationship between villus density and oxygen exchange [55, 56], whilst neglecting the branching component of the villous tree. Our model comprises two components of villus density: terminal villus density ($\phi_{TV}$) and a contribution from the branching component of the tree. Figure 5.3 shows the effect of varying $\phi_{TV}$ on the oxygen uptake capacity of the placenta. The relationship is non-linear, with low and high terminal villus density displaying low efficiency (in terms of oxygen uptake) with a peak in oxygen uptake at $\phi_{TV} \approx 0.15$ (average density of all villi $\approx 0.24$).

As villous tissue becomes more dense, there is an increase in the surface area available for oxygen exchange which increases oxygen uptake. However, there is also a concurrent increase in the terminal villous impedance to maternal blood flow, and so thus oxygen uptake becomes restricted once the tissue is sufficiently dense. Our model prediction of optimal total villus density is consistent with previous porous medium model [55], but lower than that predicted by the stream-tube model [56].

The effect of a central cavity

Like in Chapter 3, the model was solved using an artificially applied ‘central cavity’. In the presence of a central cavity, oxygen-rich maternal blood is clearly shown to project towards the chorionic plate, resulting in delivery of oxygen to deeper parts of the placental tissue (Figure 5.4a). As in baseline predictions, a decreasing central-toPeripheral oxygen
Figure 5.4: The effect of central cavities (CC) on oxygen uptake. (a) Normalised oxygen concentration field (colours) predicted for a placental unit with a central cavity which is relatively free of villous tissue. Oxygen concentration is normalised by oxygen concentration of maternal blood entering the IVS. The central cavity gives rise to deeper penetration by oxygen-rich maternal blood into the placental tissue. (b) Oxygen uptake rate is predicted to increase with cavity length with the increase in distribution of oxygen-rich blood in the IVS. However, when the cavity length is longer than 8 mm, oxygen uptake rate starts to decrease slightly.

gradient is predicted in the presence of a central cavity and is consistent with the relative antioxidant enzyme activity measured across the lobule [130].

Oxygen uptake increases with cavity length to an optimal length of approximately 8 mm (Figure 5.4b). We speculate that the 3.0–8.6 mm jets observed using colour Doppler ultrasound in near term placentas [123] are close to the optimal cavity length as small decreases in uptake occur beyond this level. This could be attributed to the increased maternal blood flow velocity (Figure 3.6b), which means that blood may be moving too fast for sufficient oxygen uptake to occur.

5.3.3 Parameter sensitivity

Table 5.3 shows how predictions of oxygen consumption are sensitive to each model parameter. Referring to Table 3.2, the parameters $\alpha$, $\kappa_{empty}$ and $x_v$ could not be estimated directly from the literature. The model is relatively insensitive to changes in $\alpha$ and $\kappa_{empty}$. As expected, the model is sensitive to $x_v$. Our model shows a decrease in oxygen uptake if DVs are situated closer to SAs, consistent with previous studies [55]. The predicted oxygen uptake rate is sensitive to the amount of oxygen brought into the placenta, via changes in $Q_{in}$ (brought upon by altered $P_{in}$ or SA diameter, $d$) or an altered $C_{in}$. Also, changes in fetal oxygen concentration ($C_f$), which reflect changes
in fetal metabolism, significantly influence predictions of oxygen consumption. Overall, structural parameters have the largest effect on model predictions of oxygen consumption, which is consistent with the concept of placental morphology having a major influence on function. Each major structural contributor to predicted function is addressed below.

### 5.3.4 The branching structure of non-terminal villi

In this section, the length of villous elements ($l_s$), branch angle ($\theta_b$), number of branching generations ($n_b$), and symmetry of the villous tree were varied over the same ranges used in Section 3.3.2. Plots showing the effects of these parameters on flow velocity emerging from the SA have previously been presented in Chapter 3 but are repeated again for ease of reference.

---

<table>
<thead>
<tr>
<th>Parameter</th>
<th>% change in parameter</th>
<th>% change in O$_2$ uptake with decrease in parameter</th>
<th>% change in O$_2$ uptake with increase in parameter</th>
</tr>
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<tr>
<td>$\tau$</td>
<td>10</td>
<td>-58.1</td>
<td>6.59</td>
</tr>
<tr>
<td>$\epsilon$</td>
<td>25</td>
<td>20.5</td>
<td>119</td>
</tr>
<tr>
<td>$x_v$</td>
<td>25</td>
<td>-24.6</td>
<td>-</td>
</tr>
<tr>
<td>$d$</td>
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<td>45.8</td>
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<tr>
<td>$l_s$</td>
<td>10</td>
<td>41.2</td>
<td>-53.1</td>
</tr>
<tr>
<td>$d_s$</td>
<td>10</td>
<td>-54.1</td>
<td>-1.83</td>
</tr>
<tr>
<td>$l_d/l_p$</td>
<td>10</td>
<td>-</td>
<td>80.0</td>
</tr>
<tr>
<td>$d_d/d_p$</td>
<td>10</td>
<td>-55.4</td>
<td>-55.5</td>
</tr>
<tr>
<td>$\theta_b$</td>
<td>10</td>
<td>-8.65</td>
<td>-8.16</td>
</tr>
<tr>
<td>$\phi_{TV}$</td>
<td>10</td>
<td>15.0</td>
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</tr>
<tr>
<td>$d_{TV}$</td>
<td>10</td>
<td>21.3</td>
<td>-16.6</td>
</tr>
</tbody>
</table>

**Maternal blood flow model**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>% change in parameter</th>
<th>% change in O$_2$ uptake with decrease in parameter</th>
<th>% change in O$_2$ uptake with increase in parameter</th>
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<tr>
<td>$\mu$</td>
<td>10</td>
<td>9.75</td>
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</tr>
<tr>
<td>$P_{in}$</td>
<td>10</td>
<td>-9.01</td>
<td>8.79</td>
</tr>
<tr>
<td>$P_{out}$</td>
<td>10</td>
<td>$1.76 \times 10^{-4}$</td>
<td>$-1.66 \times 10^{-4}$</td>
</tr>
<tr>
<td>$\kappa_{empty}$</td>
<td>10</td>
<td>-0.16</td>
<td>0.012</td>
</tr>
</tbody>
</table>

**Oxygen transport model**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>% change in parameter</th>
<th>% change in O$_2$ uptake with decrease in parameter</th>
<th>% change in O$_2$ uptake with increase in parameter</th>
</tr>
</thead>
<tbody>
<tr>
<td>$D_{O_2}$</td>
<td>10</td>
<td>$2.94 \times 10^{-4}$</td>
<td>$-2.93 \times 10^{-4}$</td>
</tr>
<tr>
<td>$C_f$</td>
<td>10</td>
<td>11.4</td>
<td>-11.4</td>
</tr>
<tr>
<td>$\alpha$</td>
<td>10</td>
<td>-1.20</td>
<td>0.99</td>
</tr>
<tr>
<td>$C_{in}$</td>
<td>10</td>
<td>-21.4</td>
<td>21.4</td>
</tr>
</tbody>
</table>
Figure 5.5: The effect of the length of the stem branch ($l_s$) on oxygen transport model predictions. The dotted line indicates the nominal value of $l_s$. (a) Oxygen uptake rate is a non-linear function of $l_s$. (b) An increase in $l_s$ gives rise to reduced number of terminal villi as the tree has fewer generations. (c) The amount of sampling grid elements containing villous tissue increases with $l_s$ at first and then begins to slowly decrease as the tree becomes more sparse. (d) Blood flow velocity into the IVS varies non-linearly with $l_s$ with similar changes to oxygen uptake rate.

**Length of villous elements ($l_s$)**

The effect of $l_s$ on placental oxygen uptake, the number of terminal elements in the branching tree, the amount of IVS filled with tissue (defined as the proportion of sampling grid elements containing branches or TBs), and velocity of blood emerging from the SA are shown in Figure 5.5. There is a non-linear relationship between $l_s$ and oxygen uptake rate. An illustration of the space-filling properties and the corresponding oxygen penetration for $l_s = 2$ mm and $l_s = 3$ mm are provided as examples in Figure 5.6. As $l_s$ increases from 1.6 mm, the villous tree begins to fill more of the IVS, enabling increased uptake of oxygen. However, as $l_s$ continues to increase, there is a reduction in the number of villous branches that can fit into the domain, resulting in a reduction in the number of terminal villi and consequently, oxygen uptake. Finally, the tree becomes sparse enough for an increase in blood flow into the IVS which brings more oxygen-rich maternal blood travelling into the IVS and oxygen uptake begins to increase again.
5.3. Results

Figure 5.6: Space-filling properties and normalised oxygen distributions predicted for villous trees with $l_s = 2$ mm and $l_s = 3$ mm. Oxygen concentration is normalised by oxygen concentration of maternal blood entering the IVS. With the increase in $l_s$, fewer longer villous branches and larger empty spaces between the branches allow further penetration of oxygen into the placental tissue.

Branch angle ($\theta_b$)

Figure 5.7 shows the effects of varying $\theta_b$ (while holding all other parameters constant) on placental oxygen uptake, the number of terminal elements in the branching tree, the amount of IVS filled with tissue, and velocity of blood emerging from the SA. Oxygen uptake rate varies non-linearly with $\theta_b$ with a peak at $\theta_b = 24^\circ$. The peaks in oxygen uptake correspond with the peaks in velocity of blood flow emerging from the SA. Even though there are only small variations in the number of terminal branches and the space filling properties of the trees with $\theta_b$, the different orientations of branches give rise to different conductivity fields. As illustrated in Figure 5.8, oxygen-rich maternal blood is able to flow more freely into the IVS for oxygen uptake as the villous branches and TBs spread out more around the inlet with the increase in $\theta_b$. However, at some point ($\theta_b \geq 26^\circ$), flow becomes shunted directly from the SA to the DVs, and bypasses the depth of villous tissue as explained in Section 3.3.2. Oxygen uptake is hence reduced as maternal blood tends to flow along this arteriovenous shunt without penetrating deep into the placental tissue for oxygen exchange.
Chapter 5. Predicting oxygenation in a placental subunit

Figure 5.7: The effect of branch angle ($\theta_b$) on oxygen transport model predictions. The dotted line indicates the nominal value of $\theta_b$. (a) Oxygen uptake rate is a non-linear function of $\theta_b$ with a peak at $\theta_b = 24^\circ$. Compared with other geometric parameters, varying $\theta_b$ results in smaller variations in (b) the number of terminal branches, and (c) the space filling properties of the generated tree. (d) Peaks in oxygen uptake correspond with peaks in blood flow velocity.
[Image: Figure 5.8: Space filling properties and normalised oxygen distributions predicted for villous trees with $\theta_b = 18^\circ$, $\theta_b = 24^\circ$ and $\theta_b = 26^\circ$. Oxygen concentration is normalised by oxygen concentration of maternal blood entering the IVS. Regions around the SA become less filled with villous tissue with increasing $\theta_b$. Flow penetrates deeper into the placental tissue, leading to an increase in oxygen uptake. Beyond $\theta_b = 26^\circ$, the large spacings between branches generate paths of high hydraulic conductivity between the SA and DVs, resulting in arteriovenous shunting.]
The effects of the number of branching generations ($n_b$) on placental oxygen uptake, the number of terminal elements in the branching tree, the amount of IVS filled with tissue, and velocity of blood emerging from the SA are shown in Figure 5.9. While $n_b$ influences the ‘filling’ of the placenta with tissue (as seen by increases in terminal elements and non-empty grid elements) (Figure 5.10), this parameter has only a small effect on flow velocity and oxygen delivery in the absence of concurrent changes in branch length or angle due to the similarity in tissue impedance around the SA.

Random variability and asymmetry

The same asymmetric tree used in Section 3.3.2 was used to predict oxygen uptake and distribution. The predicted oxygen uptake rate is close to the baseline case (6.17 mm$^3$/s (0.37 ml/min) per placental subunit), but the resulting oxygen distribution as shown
5.3. Results

Figure 5.10: Space filling properties and normalised oxygen distributions predicted for villous trees with $n_b = 12$ and $n_b = 15$. Oxygen concentration is normalised by oxygen concentration of maternal blood entering the IVS. Villous branches and TBs are oriented in similar manner near the SA for both cases, providing similar resistance to inlet flow even though the streamline patterns are different.

Figure 5.11: Normalised oxygen distribution predicted for a placental subunit containing an asymmetric villous tree with random branch angles. Oxygen concentration is normalised by oxygen concentration of maternal blood entering the IVS. Oxygen distribution is asymmetric and corresponds with the asymmetric branching of the villous tree.
in Figure 5.11 differs from the baseline case and corresponds to an asymmetric pattern based on the hydraulic conductivity of the villous tree.

Optimality of uptake with variation in geometric parameters

A peak in oxygen exchange is seen with branch lengths near to normal values (estimated from anatomical studies). The model predicts that for the same placenta size, a larger number of short villous branches result in high density tissue and obstruct maternal flow, while fewer long branches result in unimpeded maternal blood circulation but smaller gas exchange surface. The model also predicts an optimum oxygen exchange when the branch angle is $\theta_b = 24^\circ$, where villous branches and TBs are spread out sufficiently to channel maternal blood flow into the depth of the placental tissue for oxygen exchange without being shunted directly into the DVs. Without concurrent change in the branch length and angles, the number of branching generations has a small effect on oxygen exchange. The remaining model parameters, including the dimensions of the domain and maternal vasculature, blood flow and oxygen transport parameters, do not display peaks in predicted oxygen uptake within the physiological ranges analysed, although as shown in Table 5.3, some parameters show uniform increases or decreases in predicted oxygen uptake across the ranges considered.

5.3.5 The distribution of exchange interfaces

To better reflect the \textit{in vivo} distribution of oxygen uptake described in the literature [12], the distribution of oxygen exchange surfaces was varied regionally. First, the oxygen exchange capacity (or the parameter $\alpha$) was assumed to be the same at each placenta-IVS interface, but oxygen exchange was switched on and off regionally. Oxygen exchange was assumed to occur: (1) evenly through the whole IVS (no influence of structure as in previous studies [55]), (2) only in terminal villi, (3) only in stem and intermediate villi. Second, the parameter $\alpha$ was assumed to vary regionally according to the structural properties of villi and uptake was assumed to be (1) proportional to the average pO$_2$ in the regions marked in Figure 5.2a, (2) inversely proportional to the average pO$_2$ in the regions, and (3) inversely proportional to the average harmonic thickness of the villous membranes ($\tau_{vm}$) estimated by Critchley et al. [223] in the regions. Model predictions for oxygen uptake over the whole placentome and average uptake per exchange interface are shown in Figure 5.12a.
5.3. Results

(a) Oxygen uptake with different distributions of exchange interfaces compared to the baseline case where uniform uptake occurs across the whole villous tree. Oxygen exchange at non-terminal villi only resulted in a larger decrease from the baseline case than when uptake occurs at the terminal villi only. Oxygen uptake per interface is highest when exchange occurs at the terminal villi only, suggesting that the terminal villi are the main sites for efficient oxygen exchange. A small increase in oxygen uptake from the baseline case is predicted when $\alpha$ is proportional to $pO_2$. (b) Compared to constant uptake at all exchange interfaces, the model predicts the highest oxygen uptake when $\alpha$ is inversely proportional to $pO_2$ when a central cavity is present.

Figure 5.12: The effects of distributions of exchange interfaces on oxygen uptake.
Compared with the baseline case when oxygen exchange occurs across the whole villous tree, uptake by stem and intermediate villi alone resulted in a 24.5% decrease in total oxygen uptake. However, when uptake was assumed at terminal villi alone, only a 4.2% decrease was observed. In conjunction with the finding that oxygen uptake rate per exchange surface is highest ($2.97 \times 10^{-2}$ mm$^3$/s (1.78$\times10^{-3}$ ml/min)) when only terminal villi acted as exchange interfaces, our model suggests that the terminal villi of the placenta are the main sites of oxygen exchange in an efficient placenta. The model predicts an increase in oxygen uptake from the baseline case only when $\alpha$ is proportional to regional pO$_2$, or when uptake is ‘easier’ in high oxygen regions. This implies that a placental lobule functions more efficiently when uptake occurs in regions with high oxygen content (i.e. central regions). However, changes in uptake rate are small (less than 1%).

When a central cavity is present, the model predicted that highest oxygen uptake occurs when $\alpha$ is inversely proportional to pO$_2$, with a 1.9% increase in oxygen uptake when compared to constant uptake at all exchange interfaces (Figure 5.12b). This suggests that when central cavities are present, the high uptake capacity in low oxygen regions of the placentome may be optimal.

### 5.3.6 Diffusive and advective transport

Using an average pore size of 80 µm [56] as the characteristic length of the flow domain and the local speed of maternal flow at each node of the oxygen transport mesh, the spatial variation of Péclet number was estimated across the placental subunit under baseline conditions. While oxygen transport by diffusion was accounted for in all simulations through the use of the advection-diffusion-reaction equation, oxygen transport was found to be dominated by advection throughout the domain, with Péclet number less than 1 detected in the peripheral regions of the placentome near to the chorionic plate. Oxygen exchange in these subchorial regions is negligible as these regions correspond to the relatively villus-free subchorial lake and are supplied by oxygen-depleted maternal blood which has already passed through the oxygen exchange zone. Our model includes oxygen diffusion in Equation 5.1, which was neglected in previous porous medium models [55], and this analysis suggests that inclusion of oxygen diffusion is indeed important regionally.
5.4 Discussion

In this study, we have developed a model of oxygen transport in a placental subunit which attempts to incorporate the different structural characteristics of the villous tree from stem villi to terminal villi to provide a representative anatomically based description of how placental structures relate to their oxygen exchange function. Even though villous tree properties have been associated with different pregnancy outcomes [14, 16, 18, 186], it is unclear which geometrical features of the villous tree are most important for placental function. Here, our model provides an improved approach for the identification of key anatomical features that are crucial for placental exchange. Although the model is presented here in 2D, the composite approach employed is extendable to 3D as will be shown in Chapter 6, allowing a pragmatic approach which is extendable to represent the whole placenta without excessive computation, and customisable to incorporate individual placental structures from high resolution imaging techniques [123, 163, 186, 232].

Consistency with previous studies

Our model predictions compare well to whole organ measures in terms of oxygen uptake rate and the average pO$_2$ in the IVS. The oxygen distribution generated by the model also agrees with the decreasing central-to-peripheral oxygen gradient observed in primates [128, 129] and hypothesised in humans [104], and is also reflective of the oxygen distribution visualised in humans using BOLD [131].

The effect of villous branches

With the inclusion of detailed descriptions of the branching of the villous tree between the chorionic plate and basal plate in an oxygen transport model, the significant role of the branching structure of the villous tree in placental oxygen exchange can now be studied. The structure of the branching component of the villous tree (the stem and intermediate villi) plays conflicting roles in determining oxygen consumption. As demonstrated in Section 3.3.2, the villous branches act as a barrier to blood flow. Properties of the branching structure such as the branch angle, length and diameter of the stem and intermediate villi, all influence how maternal blood flow is constrained and channelled in the IVS through their contribution to IVS conductivity. At the same time, the stem and intermediate villi provide surface area for oxygen exchange. Increased villous branches would signify a higher surface area for exchange, but maternal flow could be impeded by denser villous tissue. In addition, the distribution of branches within
the IVS influences whether the villous tree is ‘space-filling’. Large empty spaces might be expected to allow free movement of maternal blood but could potentially reduce the number, and affect the distribution of terminal villi, which are the major sites for oxygen exchange, due to space constraint.

By changing terminal villus density alone, our model predicts an optimal oxygen uptake when the total villous tissue fraction is 0.24, which is slightly lower than the value of 0.25 predicted in the previous homogeneous porous medium model of Chernyavsky et al. [55]. However, our model incorporates a branching component for the villous tree and predicts that the optimality of oxygen uptake rate is influenced significantly by the branching structure of the villous tree, and local changes in maternal blood flow profiles due to this structure. Our model, and previous porous medium models, predict lower optimal villus densities than stream-tube models, whose predictions of optimal villus density (0.47) coincide with normal villus densities measured experimentally. In general, porous medium models appear to predict optimal villus density that corresponds to the total villus density in high-altitude pregnancies. Serov et al. [56] suggested several reasons for predictions of optimality to differ between the two classes of model, including the use of first order uptake kinetics (which assumes uptake is proportional to local solute concentration) and the lack of explicit representation of the uptake surface. As our model allows for local variation in villous structure, we are able to assess the implications of local variation in uptake surface by varying $\alpha$ regionally. However, this is only a qualitative approximation and cannot be accounted for with the accuracy possible in a stream-tube model that can explicitly describe the exchange barrier. Ultimately, validation of oxygen (or any nutrient) transport models in terms of predictions of optimality will rely on concurrent measurements of villous structure as suggested by Serov et al. [56], including artificial perfusion of ex vivo placentas or primate studies to provide higher resolution data than is possible in in vivo human studies. Studies of ex vivo perfused placentas can provide information on gas exchange under different maternal and placental perfusion conditions and oxygen levels, potentially providing validation data for both porous medium and stream-tube models. For example, introduction of different oxygen content in ex vivo perfusion can be used to assess whether first order kinetics is sufficient to describe oxygen exchange. Ex vivo perfusion studies combined with 3D structural imaging of the same placenta can also tell us more about how total exchange efficiency relates to villous structure.
5.4. Discussion

To date, all models are parameterised using villus density data from stereological analysis of 2D placental cross-sectional slides. Depending on the technique employed there may be errors in representing 3D villous structures in vivo including the lack of maternal and fetal blood flow, artefacts that can be introduced during the fixation process, and extrapolation to 3D. Post-partum analysis of histological villus density may overestimate the in vivo density [56]. Currently, there is no established method to correct for these factors and to verify the in vivo villus density. Although 3D imaging is constantly evolving, and high throughput methods to measure villous structure in 3D would improve the quality and quantity of data, at this stage it remains arguable whether the placenta is performing at its optimal capability. Data describing the spatial distribution of blood flow and oxygen, such as MRI images, can potentially be used for validating model predictions in terms of flow and oxygen distributions and heterogeneity. Our model is qualitatively consistent with BOLD MRI distributions of oxygenation [131], but quantitative comparison is limited by difficulties in separating out maternal and fetal components in MRI. In vivo structural and functional imaging are evolving to give a more complete picture of the 3D placenta, with both MRI and 3D ultrasound potentially providing significant new insights into blood flow rates and distributions in the placenta. Combining this type of imaging with ex vivo data in the same placentas (e.g. perfusion studies, histology, or high resolution structural imaging), provides longer-term prospects for validation of models of the placenta such as this one that predicts spatial variations of flow and oxygenation.

The effect of exchange potential

Simulations assessing the effects of exchange potential within the villous tree were conducted. When there was no distinction between terminal and stem villi in terms of exchange barrier properties, oxygen exchange was found to occur predominantly at the terminal villi, which is in line with the current understanding of the role played by terminal villi in oxygen exchange [12, 184]. In the model without a central cavity, when the exchange potential was varied as an inverse function of the average regional pO$_2$, the oxygen uptake efficiency is lower than when it was varied as a function of average regional pO$_2$. This implies that oxygen is taken up more efficiently in regions with high oxygen content (i.e. central regions) and the uptake interfaces in these regions should possess small harmonic barrier thickness in order to optimise oxygen uptake. However, in the study conducted by Critchley et al. [223], it was found that the mean harmonic barrier thickness is higher in the central regions as compared to the peripheral regions.
Simulations of the regional variation of exchange potential based on the results from Critchley et al. [223] also generated a lower uptake efficiency. On the other hand, when a central cavity is present, the model predicted that oxygen exchange efficiency is highest when the exchange potential was varied as an inverse function of the average regional pO$_2$, implying that optimum uptake occurs when the uptake capacity is high in low oxygen regions (i.e. peripheral regions) of the placentome. As this suggests a smaller harmonic barrier thickness in the peripheral region than in the central region which agrees with the observations made by Critchley et al [223], the model further corroborates the physiological existence of central cavities in placentas.

**Relevance to placental pathologies**

Although several stereological studies have reported morphological abnormalities of the villous tree in pathological conditions [13–16, 18], the structure-function relationship between the villus abnormalities and oxygen uptake efficiency remains hypothetical. As it is impossible to make direct measurement of the *in vivo* placenta, this model provides a channel to assess these proposed hypotheses. With the model sensitivity to villous tree geometry, the model results support the hypothesis that maldevelopment of placental villi will adversely affect placental oxygen exchange. Placentas affected by intrauterine growth restriction (IUGR) are generally smaller in volume [14, 16]. Stereological studies have revealed impoverished growth of villi. While the villous branch diameter remains unaffected [14, 16], the shorter villous branch length [16] give rise to a smaller villous volume and a smaller exchange surface area [14, 16]. The trophoblast epithelium was also found to be thicker [14]. A smaller placental volume (decreasing $\tau$ and $\epsilon$), coupled with a smaller exchange surface area and thicker trophoblast epithelium (decreasing $\alpha$) is predicted to lead to a decrease in oxygen uptake. Although a decrease in villous branch length ($l_s$) would result in an increase in oxygen uptake, the study by Krebs et al. [18] has suggested an increase in fetoplacental vascular impedance at the fetal capillary level as the capillary loops in the IUGR cases are sparse in number, generally elongated and less coiled than normal cases. The interplay of these factors are likely to contribute to an overall drop in the oxygen uptake rate. Assuming that oxygenated maternal blood enters into the IVS at a normal rate, the decrease in oxygen uptake rate would give rise to a higher average pO$_2$ in the IVS than in normal placentas, which is consistent with placental hyperoxia suggested by Kingdom et al. in IUGR placentas [143].
5.4. Discussion

Pre-eclamptic placentas exhibit different morphologies depending on stage of onset [16]. Early onset pre-eclampsia associated with IUGR shares similar villous morphologies as IUGR cases [16, 233]. Coupled with restricted entry of oxygenated maternal blood due to inadequate SA modelling in pre-eclamptic cases [234], the model predicts a lower oxygen uptake efficiency for early onset pre-eclamptic cases as compared to a normal placenta. In some studies, pre-eclamptic placentas present similar villous morphologies to a normal placenta [14, 151]. As the age of pre-eclampsia onset was not provided in these studies, these could be late onset cases which have been observed to possess normal placental function [16]. Given the similarities in villous structure with a normal placenta, the model predicts a relatively normal oxygen uptake rate. These model predictions are also in line with published data that reports delivery of smaller neonates to mothers with early onset pre-eclampsia [235–238] and normal birth weight neonates to mothers with late onset pre-eclampsia [236].

Model limitations

While we aim to generate villous tree structures that match as closely as possible to existing studies, we took a simplified approach of homogenising the ‘random’ structure of terminal villi into a tissue block given the complexities involved in representing the convoluted structure of terminal villi. With the lack of structural definition of the TBs in the model, it is difficult to estimate how oxygen diffusivity varies with pore sizes between the villous branches. Since our results suggest that diffusion is a minor driver for oxygen transport which only occurs mostly in the peripheral and subchorial regions of the placentome where oxygen uptake is negligible, it is reasonable to implement a uniform oxygen diffusivity of oxygen. In this model, the transport of oxygen in the fetal circulation was simplified as a perfect oxygen sink, where oxygen transferred from the maternal blood was assumed to be carried away immediately by the fetal circulation. While this situation does not describe the physiology perfectly, it does help tease the effects of materno-placental blood out from those of feto-placental blood flow to help elucidate the contributions of maternal blood to placental function.

Like earlier model of Chernyavsky et al. [55], we also simplified oxygen uptake with a first order uptake kinetic by assuming that the oxygen-haemoglobin dissociation curve is linear within the pO$_2$ range considered in the model. In future work, the uptake kinetics could be updated to reflect the non-linear oxygen-haemoglobin dissociation behaviour for a more accurate representation of oxygen exchange.
The parameter $\alpha$ is a measure of the diffusing capacity of the placenta, with the resistance across the villous membrane accounting for majority of the total diffusing capacity [133]. The resistance of villous membrane varies spatially even within the terminal villi region due to the presence of locally distended fetal capillaries which give rise to vasculosyncyntial membrane with particularly thin exchange barrier. As this distribution of vasculosyncyntial membrane was only approximated by estimating the mean harmonic barrier thickness in the different regions of the IVS, in the future the model could be adapted to incorporate specific locations of vasculosyncyntial membrane to assess their influence on uptake efficiency.

5.5 Summary and conclusions

We presented an oxygen exchange model which incorporated key geometric features of the placental villous tree. Unlike earlier models which focussed on exchange at the terminal villus level or assumed a simplified homogenous geometry for the villous tree, the model captures the different structural characteristics of the villous tree and most importantly, identifies the crucial role played by the branching structure of the villous tree in efficient oxygen exchange. Owing to the difficulties in extrapolating animal models to the human placenta and the impossibility to perform invasive experiments on human placenta during the course of pregnancy, this model can be applied to bridge the gaps in the current knowledge and postulations derived from this model can provide useful tools for understanding the structure and function relationship in normal and pathological placentas. Coupled with the development of multiscale models of placental and uterine perfusion, and combined with quantitative assessments of the anatomical structure in the human placenta, we anticipate further insights into placental physiology through pregnancy.

In the next chapter, we take a step towards a more realistic anatomical representation of the placenta. We will extend our placental subunit to a section containing multiple villous trees which are supplied by an SA distribution obtained from a human placental bed specimen, and model maternal blood flow and oxygen exchange on an organ level.
Chapter 6

Towards predicting placental oxygenation on an organ level

6.1 Motivation

In Chapters 3–5, the placenta was modelled at the level of a functional unit, where each unit comprises a single villous tree. Different distributions and orientations of SAs and DVs with respect to the villous trees have been suggested. As discussed in Section 2.3.2, some studies suggested that the villous tree is supplied by a central SA \([102–107]\), while other studies suggested SAs at the periphery of each villous tree \([110, 111]\), or a random distribution of SAs over multiple villous trees \([87, 88, 108, 109]\). It is also unclear whether the DVs are distributed randomly \([65]\) or located at the periphery of each villous tree near the septa \([66, 101, 107, 113]\). Given these uncertainties, for our models in earlier chapters we adopted the same idealised arrangement as existing computational models \([53, 55, 56, 170]\), whereby the villous tree is supplied with maternal blood entering from a central SA and leaving by two DVs near the periphery of the villous tree. However, this idealised arrangement is rarely observed in the placenta \([12]\), and the true distributions of SAs, DVs, and villous trees are likely to be more heterogeneous.

While the idealised configuration of one SA and two peripheral DVs has provided insights to the dynamics of blood flow and oxygen transport in the IVS on a subunit level, this simplified configuration may not have completely captured the structure-function of the placenta on an organ level. In Chapter 3, we established that the path of maternal blood
flow in the IVS relies on the structure of the villous tree, and is especially dependent on the orientation of the villous tree to the SA opening. Due to the contribution of villous branches and TBs to hydraulic conductivity of the IVS, maternal blood can penetrate into the IVS for gaseous exchange or totally bypass villous tissue through shunts between the SA and DVs. Although this has only been demonstrated on a placental subunit level, maternal blood flow is expected to behave in a similar manner and follow high conductivity pathways which may extend over multiple villous trees when flow is considered on an organ level. As villous trees are not completely separated from one another by the septa [112], modelling maternal blood flow in the IVS is not as straightforward as assembling the flow path obtained from independent functional units as though maternal blood flow is limited to the space of each unit.

To date, no computational model of placental blood flow or oxygen transport has been able to address the complex flow interaction arising from multiple functional units on an organ level. To bridge this gap, we extend our maternal blood flow and oxygen transport models to a placental section made up of multiple functional units. In this model, we attempt to inject anatomical realism by replacing the idealised one SA and two DVs to a single villous tree configuration with an anatomically realistic distribution of villous trees which are supplied by a distribution of SAs obtained from a basal plate section from a human placental specimen. We aim to use this 3D organ model to examine the nature of maternal blood flow and oxygen transport in a placental section made up of multiple functional units to demonstrate that maternal blood flow and oxygen transport on an organ level are not linear combinations of independent functional units. This modelling framework is parameterised with existing available data but has the potential to be customised with more structural details when high resolution imaging data becomes available.

6.2 Methods

We implemented the oxygen transport model from Chapter 5 in 3D. Implementation in 3D did not require modification of the governing equations and followed the same solution algorithm shown in Figure 5.1. However, the villous tree branching algorithm was performed in 3D and volume fraction was used in the determination of the hydraulic conductivity field (Equation 3.3). Instead of a single functional unit, we modelled a placental section comprising of multiple villous trees. To do so, we used a placental
bed specimen (obtained from a cesarean hysterectomy) from a study performed by Brosens [87]. Using the SA distribution observed on the specimen (Figure 6.1a), multiple functional units were defined in the placental section with each functional unit containing a villous tree under an SA, as detailed below.

6.2.1 Model geometry

Construction of a domain

Figure 6.1b illustrates a 3D hexahedral domain which represents the placental section. The domain has a length \( \eta \), a width \( \epsilon \), and a uniform thickness \( \tau \). The domain has \( n_{SA} \) SAs and \( n_{DV} \) DVs on the basal surface of the domain. We assumed that the SAs and DVs are \( d \times d \) square surfaces. The location of SAs in Figure 6.1a were digitised using a graph digitiser software (GraphClick). The digitised locations were used as midpoint coordinates of the SAs for generating square surfaces representative of SAs on the basal surface of the domain. In cases where boundaries of the generated SA overlap other SAs, these closely spaced SAs are represented as one SA. Figure 6.1b shows that the SA distribution used in the model corresponds to their locations on the placental specimen from Brosens [87]. As there is currently no quantitative data of DV distribution, we assumed the same number of \( n_{DV} \) as \( n_{SA} \), and randomly distributed the DVs on the basal surface of the domain (Figure 6.1b). The midpoint coordinates of each DV was derived using the RAND function in Matlab. Constraints were applied to prevent DV boundaries from overlapping with the boundaries of other SAs and DVs.

Representation of villous trees

Following the observations from earlier studies [102–107] and existing models [53, 55, 56, 170], we assumed that each villous tree is supplied by a single SA. A villous tree stem was positioned directly under the midpoint of each SA on the chorionic surface of the domain (Figure 6.1b). Seedpoints were distributed evenly throughout the domain and the branching component of each villous tree was generated in 3D with the branching algorithm described in Section 3.2.1, with the terminal convolutes homogenised as TBs.
Figure 6.1: Distribution of SAs on a placental bed section and the 3D model representation of the same section. (a) Plan view of the section shows random distribution of SAs on a placental bed from the study by Brosens [87]. Location of SAs are marked as red ‘o’ and septa insertions are denoted by black bold lines. (b) The section was represented by a hexahedral domain of thickness $\tau$, width $\epsilon$, and length $\eta$. The domain contains multiple villous trees, each generated from realistic morphometric parameters. The locations of the stem of the villous trees are marked as ‘o’ on the chorionic surface of the domain. Each villous tree is located directly under an SA (red square). Maternal blood leaves the IVS through randomly distributed DVs (blue squares).
6.2.2 Maternal blood flow in the IVS

**Governing equations and solution procedure**

Like the 2D model, maternal blood flow in the placental section was modelled as Darcy’s flow (Equation 3.2). The MFEM described in Section 3.2.2 and Appendix A was applied in 3D to solve for the velocity field and pressure distribution of blood flow in the IVS. Here, instead of area fraction, the volume fraction occupied by the villous branches or TBs was used as $\phi$ in the Kozeny-Carmen formula (Equation 3.3) to determine the spatially varying conductivity field ($\kappa(x, y, z)$). The element size of the sampling grid and the maternal flow mesh used in the solution procedure was $1 \text{ mm} \times 1 \text{ mm} \times 1 \text{ mm}$.

**Boundary conditions**

Due to the asymmetric $\kappa$ field in the domain, flow velocity ($Q_{in}$) was applied as a boundary condition at every SA element edge. Like the 2D model, the basal and chorionic surfaces act as barriers to flow, so the normal component of velocity at these surfaces were set to zero ($U_y(x, 0, z) = U_y(x, \tau, z) = 0$) except at the SAs and DVs. At the boundaries of the placental section ($x = 0$, $x = \epsilon$, $z = 0$, and $z = \eta$), a symmetry condition was applied ($U(0, y, z) = U(\epsilon, y, z)$ and $U(x, y, 0) = U(x, y, \eta)$).

6.2.3 Modelling oxygen transport

**Governing equations and solution procedure**

Oxygen transport in the placental section was represented with the advection-diffusion-reaction equation (Equation 5.1) in 3D. The same solution procedure described in Section 5.2.2 was applied to solve for oxygen distribution in the IVS using the Lagrange-Galerkin finite element method. A spatial mesh with the same resolution as the maternal flow mesh was used. A timestep of 0.005 s was implemented to solve Equation 5.1 to a steady state.

**Boundary conditions**

The oxygen concentration of maternal blood entering the domain through all SA was fixed as a constant ($C_{in}$). At all other boundaries, a zero diffusive flux was imposed ($-D_{O_2} \nabla(C_m).n = 0$, where $n$ is the outward facing normal to the boundary). Oxygen was assumed to be carried across the boundary at the DVs via the advective component of the solution.
Model outputs

The model generated the flow streamlines and pressure distributions of maternal blood, the rate of oxygen uptake by the villous trees and the distribution of oxygen concentration in the IVS. The rate of oxygen uptake by the villous trees and the average oxygen content in the IVS were calculated with the same method described in Section 5.2.2.

6.2.4 Model parameterisation

The model was parameterised to a term placenta as described in Chapter 5. Although most of the model parameters followed the same nominal values used in the 2D model, we have included Table 6.1 which lists the nominal model parameters used in this model and the physiological ranges from the literature for clarity. The length ($\eta$) and width ($\epsilon$) of the domain were based on the dimension of the placental bed specimen from Brosens’s study (Figure 6.1a) \[87\]. A 1 mm margin was added to the specimen to ensure that the SAs were within the domain boundaries. As thickness of the specimen was not provided in Brosens’s study, we adopted the same domain thickness ($\tau$) applied in the 2D model.

The same nominal values used in the 2D model were applied for parameters $l_s$, $d_s$, $d_d/d_p$, $\phi_{TV}$, and $d_{TV}$ to generate villous trees representative of a term placenta. To generate the branching component in 3D, we evenly distributed 53580 seedpoints (1 seedpoint per $1 \text{ mm}^3$) in the domain. This seedpoint density corresponds to the density required to grow a villous tree with 15 branching generations ($\sim$32000 seedpoints) if the 2D placental subunit model was extended to a $40 \text{ mm} \times 20 \text{ mm} \times 40 \text{ mm}$ hexahedral domain. $l_{min}$, $\theta_{max}$ and $f_{dist}$ were chosen to generate villous trees with an average Strahler branching ratio that falls within the range of 2.19–2.83 reported in the literature \[43\]. The key metrics describing properties of the villous trees generated for the model using the nominal parameters listed in Table 6.1 are shown in Table 6.2.

As oxygen uptake constant ($\alpha$) for the placental section cannot be estimated directly from the literature, we assigned an arbitrary constant of $0.083 \text{ s}^{-1}$ for $\alpha$ and this value can be fine-tuned when data becomes available. Except for $\alpha$, other parameters related to the maternal blood flow model and oxygen transport model followed the same nominal values used in the 2D model and discussion of how the values were selected for these parameters is found in Sections 3.2.3 and 5.2.3.
Table 6.1: Model parameters for a 3D term placental section. The table also includes chosen nominal value and literature range for each parameter.

<table>
<thead>
<tr>
<th>Symbol</th>
<th>Description</th>
<th>Nominal value</th>
<th>Literature range</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Model Geometry</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>$\tau$</td>
<td>Domain thickness</td>
<td>20 mm</td>
<td>20–45 mm</td>
<td>[12, 55, 65, 182]</td>
</tr>
<tr>
<td>$\varepsilon$</td>
<td>Domain width</td>
<td>47 mm</td>
<td>45 mm</td>
<td>[87]</td>
</tr>
<tr>
<td>$\eta$</td>
<td>Domain length</td>
<td>57 mm</td>
<td>55 mm</td>
<td>[87]</td>
</tr>
<tr>
<td>$n_{SA}$</td>
<td>Number of SAs</td>
<td>31</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td>$n_{DV}$</td>
<td>Number of DVs</td>
<td>31</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td>$n_{villous}$</td>
<td>Number of villous trees</td>
<td>31</td>
<td>40–70 (whole placenta)</td>
<td>[61, 65, 66, 72]</td>
</tr>
<tr>
<td>$d$</td>
<td>Diameter of SAs and DVs</td>
<td>2 mm</td>
<td>2–3 mm</td>
<td>[104, 185]</td>
</tr>
<tr>
<td>$n_{seed}$</td>
<td>Number of seedpoints</td>
<td>53580</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td>$l_s$</td>
<td>Stem length</td>
<td>2 mm</td>
<td>2 mm</td>
<td>[39, 63]</td>
</tr>
<tr>
<td>$d_s$</td>
<td>Stem diameter</td>
<td>1.70 mm</td>
<td>1.70 mm</td>
<td>[63]</td>
</tr>
<tr>
<td>$l_{min}$</td>
<td>Minimum branch length</td>
<td>0.01 mm</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td>$\theta_{max}$</td>
<td>Maximum branch angle</td>
<td>20$^\circ$</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td>$f_{dist}$</td>
<td>Fractional distance to centre of mass</td>
<td>0.40</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td>$d_d/d_p$</td>
<td>Daughter/parent branch diameter ratio</td>
<td>0.80</td>
<td>~0.80</td>
<td>[63]</td>
</tr>
<tr>
<td>$\phi_{TV}$</td>
<td>Terminal villi area fraction</td>
<td>0.40</td>
<td>0.28–0.40</td>
<td>[14, 16]</td>
</tr>
<tr>
<td>$d_{TV}$</td>
<td>Terminal villi diameter</td>
<td>0.05 mm</td>
<td>0.03–0.06 mm</td>
<td>[16, 63]</td>
</tr>
<tr>
<td>Maternal blood flow model</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>$\mu$</td>
<td>Blood viscosity</td>
<td>$4.00 \times 10^{-3}$ Pa.s</td>
<td>$4.00 \times 10^{-3}$ Pa.s</td>
<td>[55]</td>
</tr>
<tr>
<td>$Q_{in}$</td>
<td>Inlet flow velocity</td>
<td>83.3 mm$^3$/s (5 ml/min)</td>
<td>83.3–125 mm$^3$/s (5–7.5 ml/min)</td>
<td>[55, 92–95]</td>
</tr>
<tr>
<td>$\kappa_{empty}$</td>
<td>Maximum hydraulic conductivity</td>
<td>0.52 mm$^2$</td>
<td>$\infty$</td>
<td></td>
</tr>
<tr>
<td>Oxygen exchange model</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>$D_{O_2}$</td>
<td>Diffusivity of oxygen in blood</td>
<td>$1.62 \times 10^{-4}$ mm$^2$/s</td>
<td>$1.62 \times 10^{-4}$ mm$^2$/s</td>
<td>[224]</td>
</tr>
<tr>
<td>$C_f$</td>
<td>Oxygen concentration in fetal blood</td>
<td>3.43 mol/m$^3$</td>
<td>3.43–8.17 mol/m$^3$</td>
<td>[225]</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(8.72$\times 10^{-2}$ ml/ml)</td>
<td>(8.72$\times 10^{-2}$–2.08$\times 10^{-1}$ ml/ml)</td>
<td></td>
</tr>
<tr>
<td>$\alpha$</td>
<td>Oxygen uptake constant</td>
<td>0.083 s$^{-1}$</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td>$C_{in}$</td>
<td>Oxygen concentration in SA</td>
<td>6.44 mol/m$^3$</td>
<td>6.40–6.52 mol/m$^3$</td>
<td>Appendix D, [226]</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(1.64$\times 10^{-1}$ ml/ml)</td>
<td>(1.63$\times 10^{-1}$–1.66$\times 10^{-1}$ ml/ml)</td>
<td></td>
</tr>
</tbody>
</table>
Table 6.2: Key metrics of villous trees generated for a placental section at term using nominal parameters from Table 6.1.

<table>
<thead>
<tr>
<th>Metric</th>
<th>Generated Value (s.d.)</th>
<th>Literature value when available [References]</th>
</tr>
</thead>
<tbody>
<tr>
<td>Maximum generations</td>
<td>19</td>
<td>7–34 [12, 62–64]</td>
</tr>
<tr>
<td>Mean number of generations</td>
<td>12</td>
<td>15 [63]</td>
</tr>
<tr>
<td>Mean branch angle</td>
<td>48.9° (28.3°)</td>
<td>–</td>
</tr>
<tr>
<td>Major/minor branch angles</td>
<td>0.98</td>
<td>–</td>
</tr>
<tr>
<td>Strahler branching ratio</td>
<td>2.55</td>
<td>2.19–2.83 [43]</td>
</tr>
<tr>
<td>Diameter of terminal vessel branches</td>
<td>0.03 mm</td>
<td>–</td>
</tr>
</tbody>
</table>

6.3 Results

6.3.1 Distribution of villous trees

Using nominal parameter values, the model generated 31 non-uniformly distributed asymmetric villous trees. The villous trees are closely packed together in the placental section, with some villous branches interlacing with branches from neighbouring villous trees (Figure 6.2a). As shown in Figure 6.2b, the villous trees assume different shapes and sizes and in some portions, they are separated by empty gaps which roughly follow the septa insertion pattern recorded by Brosens [87] (black lines in Figure 6.1a). The generated villous trees also bear a branching structure that is similar to previously generated placental vasculature trees [168]. This is qualitatively similar to the structures obtained from corrosion casts of fetal cotyledons [121], an example of which is shown in Figure 6.2c, but currently few quantitative metrics for comparison exist in the literature.

6.3.2 Flow streamlines

Figure 6.3 shows different views of the streamlines which define maternal blood flow from the SAs to the DVs. Some DVs receive maternal blood from more than one SA. The model predicts that maternal blood entering the IVS from the SAs is not restricted to the space of the villous tree beneath the SA but traverses across multiple villous trees in some instances. Also, maternal blood from the SAs does not necessarily always drain through the closest DV. When viewed from the $x$-$y$ and $y$-$z$ planes (Figures 6.3c and d),
6.3. Results

Figure 6.2: Model representations of villous trees distribution. (a) Perspective view of the model domain shows densely packed villous trees with villous branches interlacing branches from neighbouring villous trees. (b) Plan view of the generated villous trees (villous branches are represented with lines) reveals a similar pattern between the empty gaps between villous trees and septa insertions observed in the placental specimen by Brosens [87]. (c) Corrosion cast of the fetal cotyledon from a human placenta [121] displays a branching structure that is qualitatively similar to the villous trees generated by the model.

the flow streamlines exhibit a range of penetration depth. Most of the streamlines do not penetrate more than half of the domain thickness, with some streamlines ‘short-circuiting’ from the SAs to their nearest DVs.

6.3.3 Oxygen uptake and distribution

The model predicts an average oxygen uptake of 4.20 mm$^3$/s (0.25 ml/min) by the placental section. Given that the volume of the placental section ($5.36 \times 10^4$ mm$^3$) is approximately 0.15 of the total volume of an average term placenta ($3.68 \times 10^5$ mm$^3$ [42]),
Chapter 6. Towards predicting placental oxygenation on an organ level

Figure 6.3: Streamlines predicted for a placental section. (a) The perspective and (b) plan views show that flow from an SA (red box) does not always drain through the nearest DV (blue box) and can traverse across several villous tree before draining through a DV. Views from the (c) $x$-$y$ and (d) $y$-$z$ planes show that the flow streamlines between the SAs and DVS have varying penetration depths. Most streamlines penetrate less than half of the thickness of the placenta, with some ‘short-circuiting’ from the SA to its closest DV.

The predicted oxygen uptake is lower than the oxygen consumption of 45–157 mm$^3$/s (2.70–9.41 ml/min) estimated for a similar sized placental section [227]. The average pO$_2$ in the IVS predicted by the model is 4200 Pa (31.5 mmHg), which is close to the upper limit of the range 2400–4520 Pa (18.0–33.9 mmHg) measured in the literature [226, 228, 229].

Oxygen distribution in the middle cross-sectional planes defined by $(x, y, 28)$ and $(23, y, z)$ are provided in Figure 6.4 for purpose of illustration. The location of these sections are marked with dotted lines in Figure 6.3b. Like in these two sections, oxygen distribution from all sections of the domain are generally heterogeneous. Oxygen content is highest near the SA inlets and decreases as oxygen is taken up by the villous trees when maternal
Figure 6.4: Normalised oxygen distributions predicted for a placental section. Oxygen concentration is normalised by oxygen concentration of maternal blood entering the IVS. Like oxygen distributions in planes (a) \((x, y, 28)\) and (b) \((23, y, z)\), all 2D sections obtained from the 3D model exhibit a higher oxygen content near SA inlets at the basal surface. Oxygen content decreases as oxygen is taken up by the villous trees when maternal blood traverses the IVS to the DVs.

This results in lower oxygen content near the chorionic plate as compared to the region near the basal plate. This distribution also corresponds well to BOLD images of 28–36 weeks normoxic placentas \([131]\). As shown in a sample BOLD image (Figure 6.5), the bright areas corresponding to regions with higher oxygen content are found along the basal surface, with a decreasing voxel intensity (decreasing oxygen content) towards the chorionic plate.

6.4 Discussion

In this chapter, a 3D model of maternal blood flow and oxygen transport is presented for a placental section made up of multiple functional units. Here, we departed from the
Figure 6.5: BOLD image of a placenta in a cross-section through uterus [131]. The placenta (outlined in red) presents a heterogeneous distribution similar to the oxygen distribution predicted by the model. Dark areas (low oxygen) are scattered near the chorionic side (white arrow) which extend towards the brighter areas (high oxygen) near the opposing basal side of the placenta.

A distribution of SAs obtained from a placental bed specimen was used in this model to provide an anatomical realistic representation of the SAs [87]. Although there have been conflicting accounts about the orientation of SA to villous tree (Section 2.3.2), we followed earlier models [53, 55, 56, 170] and assumed that each villous tree is located directly under an SA opening. With the SA distribution on the placental specimen [87], the model generated asymmetric villous trees that exhibit structural resemblance to the villous structures obtained from corrosion cast of fetal cotyledons. Unlike existing placental subunit models where villous tree structures are assumed to conform to the shape of the domain, individual villous trees generated by the model have non-uniform shapes and sizes throughout the domain as there are no ‘wall boundaries’ that constrain the space of each villous tree. The generated villous trees are closely packed together as...
described in the literature [121, 231], with some villous tree branches interlacing with branches from neighbouring trees. The distribution of villous trees also compares well with the placental specimen used in this study as the boundaries of generated trees roughly follow the pattern of septa insertion observed in the placental sample.

With the inclusion of multiple villous trees and a random distribution of DVs, the model predicts that flow from the SAs does not always drain through the closest DV and in some cases, traverses over several villous trees before leaving the IVS through a further DV. Instead of restricting the flow path within the confines of a single placental subunit, the model captures a hydraulic conductivity field that is continuous over the villous trees within the placental section. Based on the structural properties of the villous trees, a path of highest hydraulic conductivity sometimes extends beyond more than one villous tree, resulting in the traversal of maternal blood flow across multiple villous trees as flow follows the path of highest conductivity as demonstrated previously in the context of a single placental subunit (Chapter 3). This confirms that the dynamics of blood flow and oxygen transport in the IVS are not linear combinations of single placental subunits, and motivates the need to simulate placental blood flow and oxygen transport on an organ level for a more accurate representation of the placental structure-function relationship.

The model has adequately predicted a heterogeneous distribution of oxygen as observed in BOLD images [131], where oxygen-rich regions are near the basal surface where oxygen is delivered by the SAs, and oxygen level decreases towards the chorionic surface as oxygen is taken up by the villous trees as maternal blood circulates through the IVS. However, the oxygen uptake predicted by the model falls short of the oxygen consumption estimated from whole placentas obtained from women undergoing elective cesarean section [227]. This discrepancy is mainly attributed to the data used to parameterise this model. For example, the oxygen uptake constant $\alpha$ for the placental section cannot be estimated from the literature and an increase in the arbitrary constant selected for the model potentially drives oxygen uptake by the villous trees, as previously demonstrated in the placental subunit model (Table 5.3). As the average oxygen content in the IVS (4200 Pa (31.5 mmHg)) is near the higher limit of the literature range of 2400–4520 Pa (18.0–33.9 mmHg) [226, 228, 229], there is still margin to increase $\alpha$ while maintaining an average IVS $pO_2$ within the reported range. In this study, we parameterised our maternal vessels distribution based on the placental bed specimen from Brosens’s study [87]. Although the placental bed specimen represents a two-fifth section of a whole
placenta \[87\], the total area of the placental bed estimated from the size of the section \((6.20 \times 10^3 \text{ mm}^2)\) appears smaller than the area of the placental bed of a term placenta \((3.80 \times 10^4 \text{ mm}^2)\) \[59\]. Furthermore, as data such as the mass of the placental section was not provided in Brosens’s study, we were unable to make quantitative comparison of oxygen uptake predicted by the model against measurements from whole placentas obtained from a different study \[227\].

**Model limitations**

In this model, effort has been made to reproduce an SA distribution that closely matches an actual placental specimen. Unfortunately, SAs that are in close proximity were represented as a single vessel for computational tractability. Even though a sufficiently refined spatial mesh is capable of providing a faithful representation of the SA distribution in the model, the balance between its computational cost and whether such replication would provide additional biological insights will need to be assessed before undertaking such modelling efforts in future studies.

### 6.5 Summary and conclusions

In this chapter, we extended our modelling framework to include multiple functional units. Our model is able to generate an anatomically realistic representation of a section of the placenta which comprises several villous trees which are fed by assumed or observed SA distributions. This model provides an improved approach to study how blood flow and oxygen transport are influenced by the structural characteristics of an ensemble of villous trees. When multiple villous trees are considered on the whole, maternal blood flow sometimes traverses several functional units as the path of highest conductivity extends beyond more than one villous tree. Hence, to attain a further understanding of the structure-function relationship in the placenta, existing computational models have to be taken from the placental subunit level to an organ level. In the next chapter, we summarise the findings of this thesis and propose research directions for future work.
Chapter 7

Conclusions and future research directions

7.1 Thesis summary

This thesis focuses on the development of a multiscale model of blood flow and transport (for example, of oxygen) in the human placenta. Through simulations with controlled perturbations, the model findings have advanced our current understanding of the placenta through identification of key parameters that influence placental function and elucidation of events that are happening in the placenta which cannot be observed directly through imaging or experimental measurements. Furthermore, the model also provided a predictive tool that enables hypotheses to be tested \textit{in silico} without conducting unethical or infeasible experiments on living human subjects.

Our model included the well-defined branching structure of the largest villous tree branches, as well as a smoothed representation of the small terminal villi that comprise the placenta’s gas exchange interfaces. In Chapter 3, the model demonstrated that maternal blood flow is sensitive to the branching component of the villous tree, which is missing from previous models. The model predicted that the distribution of villous branches within the IVS influences their contribution to IVS conductivity, and hence, determines the flow path for maternal blood in the IVS. The model identified a new mechanism for arteriovenous shunting, which is derived from large empty spaces between
the villous branches that allow maternal blood to bypass the villous tree and drain directly from the SAs to the DVs.

In Chapter 4, we applied our model to interpret MRI images of placental blood flow generated by the IVIM technique. This technique assumes that tissue perfusion is a quasi-diffusive process and separates the image voxel into a tissue compartment undergoing molecular diffusion and a perfusion compartment with ‘pseudo-diffusion’ due to blood movement. However, it is unclear whether the same assumptions that is employed to analyse a single micro-circulatory network in tissues can be applied to analyse the two independent fetal and maternal blood flow systems in the placenta. The model demonstrated that fetal blood flow in a vessel tree exhibits pseudo-diffusive behaviour provided the vessel density and flow velocity are above the detection limit. In voxels with coherent flow (such as maternal blood flow near the SA), flow is represented by a pseudo-diffusion coefficient that decreases with flow magnitude. However, in some voxels with varying flow magnitudes and directions, IVIM generates a perfusion fraction of less than 1, even though the voxel is completely filled with moving fluid. The model suggests that while IVIM separates the voxel into two compartments, the voxel in placental images is quantified based on two representative pseudo-diffusion coefficients, rather than a simple diffusion and perfusion component as per the conventional assumption.

We then coupled our maternal blood flow model to a model of oxygen transport to investigate how structural heterogeneity of the placenta affects oxygen transport and uptake in the space around a placental functional unit (the villous tree) in Chapter 5. The model demonstrated that oxygen exchange is sensitive to villous tree geometry, including the villous branch length and volume, which are known to change in pathologies. This is because, to be an efficient exchanger, the architecture of the villous tree must provide a balance between maximising the surface area available for exchange, and the opposing condition of allowing sufficient maternal blood flow to penetrate into the space surrounding the tree. The model also predicted an optimum oxygen exchange when the branch angle is 24°, as villous branches and TBs are spread out sufficiently to channel maternal blood flow deep into the placental tissue for oxygen exchange without being shunted directly into the DVs. Without concurrent change in the branch length and angles, the model suggested that the number of branching generations has a small influence on oxygen exchange.
To further inject structural realism, the modelling framework was extended to a section of the placenta containing multiple villous trees which are fed by an SA distribution from a placental bed specimen in Chapter 6. Through this model, we demonstrated that blood flow and oxygen transport is not a linear combination of independent placental functional units, and needs to be studied on an organ level. With advancement of high-resolution imaging techniques that are currently evolving to better quantify placental structure, this research provides an organ level infrastructure that is readily customisable for different studies which are useful for advancing our understanding of the placenta.

7.2 Future work

7.2.1 Orientation between villous tree and maternal blood vessels

In Chapter 3, we established that the orientation of the villous tree with respect to the SA inlet influences maternal blood flow in the IVS. For example, a highly conductive path directly downstream of the inlet in the form of a central cavity increases the penetration depth of maternal blood, while widely spaced villous branches or TBs between the SAs and DVs give rise to arteriovenous shunts between the SA and DV. A similar relationship between the orientation of villous tree with respect to the SA and the penetration depth of maternal blood was observed using the organ model in Chapter 6. As each villous tree in the organ model has a different branching pattern, they provide varying degrees of obstruction to the incoming maternal blood, thereby generating a range of penetration depths for streamlines originating from different SAs. Although we assumed that the villous tree is located centrally downstream of an SA, this orientation is rarely observed in the whole placenta [12]. While ex vivo analysis of SA openings and villous trees would be most informative, it is difficult to obtain such data as hysterectomy of a pregnant womb is required. This drives the need to focus on methods to infer maternal blood vessels and placental structures from in vivo imaging.

In the meantime, it would be worthwhile to use the organ model to examine how different orientations of villous trees to the SA affect maternal blood flow and oxygen exchange, given the dependence of maternal blood flow on the structure of the villous tree near the SA inlet. For instance, determination of placental function as the SAs are moved from the centre of a villous tree to a peripheral location will allow us to compare the
efficiency of different SA-to-villous tree orientations that have been reported in the literature (Section 2.3.2). Some streamlines have been predicted to bypass the IVS through ‘short-circuits’ between SAs and their nearest DVs, which is undesirable for oxygen exchange. As our modelling framework incorporates a hydraulic conductivity field that varies spatially with the structural properties of the villous trees, it would be beneficial to conduct a similar study as the one performed by Chernyavsky et al. [55] to investigate how the number of DVs and their relative distance from an SA interact with the structure of the villous trees so as to tease out how these parameters can affect placental oxygen exchange efficiency on an organ level.

7.2.2 Imaging and experimental data

We anticipate improvement in model accuracy with the quality of data available for model parameterisation. Apart from incorporating details about placental branching structure from microscopic studies [163, 186, 239], an area for improvement lies in better representing the placental shape in the model. In the current modelling framework, the entire placental section has a uniform thickness. However, the villous trees in the placenta have varying heights with taller trees in the central region and shorter ones towards the periphery [59]. As the thickness of the placenta alters the hydraulic conductivity field and hence blood flow in the IVS, the height of the villous trees is an important consideration for future studies. When available, data on placental shape and volume is useful for constructing structural meshes to replace the current hexahedral domain for model solution. In addition, locations of the septa in placental samples provide information that better defines the boundaries of the neighbouring villous trees. Even though septa do not completely partition the placental lobes [112], their contribution to the hydraulic conductivity field in the IVS should be accounted for in the model since they could potentially hinder a conductive path between multiple villous trees and force maternal blood flow to follow a particular flow path in the IVS.

Ideally, the models in this thesis should be parameterised and tested with data from the same placenta. An option for model parameterisation is to collect data such as the overall placental shape and volume by performing micro-CT of a cast created from the delivered placenta, and verify these structural data against ultrasound and/or MRI reconstructed images obtained in utero. Micro-CT imaging of placental beds for SAs and
DVs distribution also provides a means for supplementing in vivo ultrasound data which identifies the location of ‘jets’ for parameterising the distribution of maternal blood vessels in the models [123]. An essential step is to adequately validate the model by comparing oxygen uptake predicted by the model against the oxygen consumption measured from the same placenta prior to delivery or through artificial perfusion experiments post-delivery. Imaging techniques such as BOLD can be applied to visualise regional distribution of oxygenation in the placenta and provide comparison to model predictions. Since voxel intensity in BOLD images represents the level of oxygenation in the organ, quantification of the proportion of voxels having an intensity above a threshold (defined to indicate an ‘oxygen-rich’ voxel) is a means for assessing heterogeneity in oxygenation in the placenta. The same analysis could be performed on the oxygen distribution predicted by the model through sampling the distribution with elements having a size close to the dimension of the voxel used in the analysis of BOLD images. A threshold oxygen content that corresponds to the threshold intensity could be applied to determine whether the model predicts a similar proportion of voxels above the threshold oxygen content. Another method to quantify heterogeneity in oxygenation is to calculate a coefficient of variation\footnote{Coefficient of variation is calculated by dividing the standard deviation of voxel value by the mean voxel value.} from the intensity of voxels in BOLD images and the average oxygen content in the sampling elements of the oxygen distribution predicted by the model.

### 7.2.3 Pathological placentas

Apart from normal placentas, future work involves customising the model to pathological placentas, by including structural abnormalities such as incomplete remodelling of the SAs in pre-eclamptic placentas [76], or reduced terminal villus volume [14, 16], to predict the oxygen exchange efficiency in pathological placentas. Not only does the current modelling framework provide a platform to understand how structural abnormalities affect oxygen exchange efficiency in the placenta, in the long run, the model holds potential to be developed into a tool for predicting pregnancy outcomes based on the structural features observed during pregnancy through placental imaging.
7.3 Final words

In this thesis, we developed a framework to incorporate heterogeneous structural features of the placenta (e.g. villous branches) to model IVS blood flow and gaseous exchange. We built the modelling framework from simple 2D representations of a placental unit, within which we could assess key structural parameters, and ultimately extended our modelling framework to include multiple functional units. Our models provide an improved approach to study how blood flow and oxygen transport are influenced by the structural characteristics of individual, as well as ensemble, villous trees. With advances in imaging technologies, we anticipate increasing accuracy in parameterisation and rigorous model validation. The modelling framework developed in this thesis thus provides a ready-to-use platform which is easily customisable for different structure-function studies, which would contribute further knowledge about normal and pathological placental physiology in the future.
Appendix A

Mixed finite element method

Here we present the mathematical formulation of the mixed finite element method (MFEM) and its application for solving Darcy’s equations [175, 178, 179].

A.1 Weak formulation

Maternal blood flow in the IVS is described by (Chapter 3, Equation 3.2):

\[
\begin{align*}
\nabla \cdot \mathbf{V} &= 0, \\
K^{-1}\mathbf{V} + \nabla P &= 0,
\end{align*}
\]  

where \( \mathbf{V} \) is the blood velocity in the IVS, \( K \) is the proportionality constant equivalent to the division of the hydraulic conductivity (\( \kappa \)) by the viscosity of maternal blood (\( \mu \)), and \( \nabla P \) is the local maternal blood pressure gradient.

The MFEM uses two individual approximation spaces for pressure and velocity fields with solutions obtained simultaneously. To do this, we derived the weak formulation for the governing equations. The weak form for Equation A.1a was obtained by applying a scalar weighting function \( \nu \) and integrating Equation A.1a over the domain \( \Omega \), while the weak formulation for Equation A.1b was obtained by imposing a vector weighting function \( \omega \).
and integrating over the domain $\Omega$, and by applying Green’s formula. Then

\begin{equation}
\int_{\Omega} \nu_E \nabla \cdot \nabla \partial \Omega = 0, \tag{A.2a}
\end{equation}

\begin{equation}
\int_{\Omega} K^{-1} \nu \omega \partial \Omega - \int_{\Omega} P \nabla \cdot \omega \partial \Omega = - \int_{\Gamma} P \omega \cdot n, \tag{A.2b}
\end{equation}

where $n$ is the outward facing normal to the domain boundary $\Gamma$.

### A.2 Approximation spaces

The pressure space was approximated with piecewise constant function,

\begin{equation}
P = \sum_{E=1}^{Ne} P_E \nu_E, \tag{A.3}
\end{equation}

where $Ne$ is the total number of elements, $E$ is the element number, $P_E$ is the pressure of element $E$, and the pressure basis function $\nu_E$ is equal to one inside an element, and zero elsewhere. This implies that each mesh element was assigned a pressure which is constant over the element, and there were as many pressure unknowns as the number of mesh elements.

The velocity space was approximated using a zero order Raviart-Thomas space (RT0) [240]. In this thesis, velocity fields in the RT0 space were used and described by a constant normal component of the velocity across mesh element edges (2D) or faces (3D):

\begin{equation}
V = \sum_{i=1}^{Ned} Q_i \omega_i, \tag{A.4}
\end{equation}

where $Ned$ is the total number of element edges (2D) or faces (3D), $i$ is the edge or surface number, and $Q_i$ corresponds to the normal component of the velocity across edge or surface $i$. The degrees of freedom of an RT0 velocity field are defined by the fluxes through each face of the element, for instance, three degrees of freedom are required for a 2D triangular element, while six degrees of freedom are necessary for a hexahedral element. Each of these degrees of freedom was associated with a shape function $\omega_i$, which is a vector field that has a unit flux through a given face and the normal component of velocity on all other faces is equal to zero as illustrated in Figures A.1a and b. This ensures a continuous normal flux across the edge or surface shared by two adjacent elements.
A.2. Approximation spaces

Figure A.1: RT0 basis function and local node numbering for 2D and 3D elements. (a) Basis function for one of the edges of the 2D triangular element and (b) one of the surfaces of the 3D hexahedral element. (c) Vertices of a 2D triangular RT0 element are numbered locally as \( P_i \) (i=1–3) in a counterclockwise direction with each edge \( E_l \) assigned the same subscript number as the opposite vertex number. (d) The local numbering convention for faces on a 3D hexahedral RT0 element with dimension \([0, \Delta x] \times [0, \Delta y] \times [0, \Delta z]\).

For a 2D triangular element \( \Delta P_1P_2P_3 \) as illustrated in Figure A.1c, the vector basis function \( (\omega_i) \) is expressed as:

\[
\omega_i = \frac{|E_l_i|}{2A_\Delta}(x - V X_i),
\]

where \(|E_l_i|\) is the length of edge \( i \), \( A_\Delta \) is the area of the reference triangular element, and \( V X_i \) is the coordinate of vertex \( P_i \) as labelled in Figure A.1c.
For a 3D hexahedral element as shown in Figure A.1d, $\omega_i$ is

\[
\begin{align*}
\omega_1 &= \frac{1}{\Delta x \Delta y \Delta z} \begin{bmatrix} x \\ 0 \\ 0 \end{bmatrix}, \\
\omega_2 &= \frac{1}{\Delta x \Delta y \Delta z} \begin{bmatrix} x - \Delta x \\ 0 \\ 0 \end{bmatrix}, \\
\omega_3 &= \frac{1}{\Delta x \Delta y \Delta z} \begin{bmatrix} 0 \\ y \\ 0 \end{bmatrix}, \\
\omega_4 &= \frac{1}{\Delta x \Delta y \Delta z} \begin{bmatrix} 0 \\ y - \Delta y \\ 0 \end{bmatrix}, \\
\omega_5 &= \frac{1}{\Delta x \Delta y \Delta z} \begin{bmatrix} 0 \\ 0 \\ z \end{bmatrix}, \\
\omega_6 &= \frac{1}{\Delta x \Delta y \Delta z} \begin{bmatrix} 0 \\ 0 \\ z - \Delta z \end{bmatrix},
\end{align*}
\]

where $\Delta x$, $\Delta y$, and $\Delta z$ are the length of the element in the $x$-, $y$-, and $z$-directions, respectively.

### A.3 Discretization of equations

Substitution of Equations A.3 and A.4 into Equation A.2 gives

\[
\begin{align*}
\sum_{i=1}^{\text{Ned}} Q_i \int_{\Omega} \nabla \cdot \omega_i \, \nu_E \, \partial \Omega &= 0, \\
\sum_{i=1}^{\text{Ned}} Q_i \int_{\Omega} (K^{-1} \omega_i) \, \omega \, \partial \Omega - \sum_{E=1}^{\text{Ne}} P_E \int_{E} \nabla \cdot \omega \, \nu_E \, \partial \Omega &= - \int_{\Gamma} P \omega \cdot \mathbf{n}.
\end{align*}
\]

As $\nu_E = 1$ over element $E$ and 0 elsewhere, Equation A.6 is re-written for each element in the domain as

\[
\begin{align*}
\sum_{k=1}^{nE} Q_k \int_{E} \nabla \cdot \omega_k \, \partial \Omega &= 0, \\
\sum_{k=1}^{nE} Q_k \int_{E} (K^{-1} \omega_k) \, \omega \, \partial \Omega - P_E \int_{E} \nabla \cdot \omega \, \partial \Omega &= -TP_k,
\end{align*}
\]

where $nE$ is the number of edges or surfaces in an element, $k$ is the local numbering for edges or faces in element $E$, and $TP_k$ is the normal pressure flux on the edges or surfaces.
As specified by \(- \int_{\Gamma} P_E \omega \cdot \mathbf{n}\). As pressure is continuous across adjacent element edges or faces, \(TP\) for interior edges or surfaces is always zero, while \(TP\) for boundary surfaces will be defined by pressure heads provided by the boundary conditions.

**A.4 Assembly of equations and boundary conditions**

Equation A.7, for all mesh elements, was assembled into a system of equations represented by:

\[
\begin{bmatrix}
A1 & -A2 \\
A2^T & 0
\end{bmatrix}
\begin{bmatrix}
Q \\
P
\end{bmatrix} =
\begin{bmatrix}
bD \\
0
\end{bmatrix},
\]

where \(A1 = \int_{\Omega} (K^{-1} \omega_i) \omega \partial \Omega\), \(A2 = \int_{\Omega} \nabla \cdot \omega \nu_E \partial \Omega\), \(A2^T\) is the transpose of \(A2\), and \(bD\) represents the Dirichlet boundary conditions, defined by known pressures at the boundary edges. \(Q\) is a vector representation of the unknown fluxes across edges or faces while \(P\) represents the unknown pressures at the elements.

Where necessary, Dirichlet boundary conditions were introduced by defining known pressures at boundary elements (for example, pressure at the inlet and outlet edge) in \(bD\). Neumann conditions were imposed by assigning a prescribed flux to the boundary edge or face, such as flux on the inlet edge or zero flux on wall surfaces. Neumann conditions were incorporated in the system of equations by replacing the corresponding flux in \(Q\) with the prescribed value before solving the equations.

**A.5 Solutions**

The MFEM generates piecewise constant pressure for each of the mesh element and velocity fluxes at each of the element edge or face. Within a 2D triangular element, a velocity field \(\mathbf{u}\) is given by:

\[
\hat{\mathbf{u}} = \begin{bmatrix} a_1 + bx \\ a_2 + by \end{bmatrix}
\]
Appendix A. Mixed finite element method

with $a_1 = \hat{u}_1$, $a_2 = \hat{u}_2$, where the $\hat{u}_1$ and $\hat{u}_2$ refer to the velocity flux on edge number 1 and 2 respectively as shown in Figure A.1c. Since the velocity field is divergence-free, $b = 0$.

The velocity field $\hat{u}$ for a 3D hexahedral element is expressed as

$$\hat{u} = \begin{bmatrix} a_1 + b_1 x \\ a_2 + b_2 y \\ a_3 + b_3 z \end{bmatrix}, \quad (A.10)$$

where

$$a_1 = \hat{u}_2, \quad b_1 = \frac{\hat{u}_1 - \hat{u}_2}{\Delta x}, \quad (A.11a)$$

$$a_2 = \hat{u}_4, \quad b_2 = \frac{\hat{u}_3 - \hat{u}_4}{\Delta y}, \quad (A.11b)$$

$$a_3 = \hat{u}_6, \quad b_3 = \frac{\hat{u}_5 - \hat{u}_6}{\Delta z}, \quad (A.11c)$$

and $\hat{u}_i$ are the normal velocities associated to face $i$ as illustrated in Figure A.1d.
Appendix B

IVIM model derivation and verification

Here we provide details on derivation of the modified Bloch equation, present results from verification and convergence analysis performed on a simple IVIM model comprising of 1D flow in a single line segment, and describe how the signal attenuation curve is fitted to a biexponential function.

B.1 Derivation of modified Bloch Equation

To derive Equation 4.11 in Section 4.2.2, Equation 4.9 is substituted with Equation 4.10. As \( \frac{\partial \mathbf{M}}{\partial t} = \frac{\partial M_x}{\partial t} \hat{i} + \frac{\partial M_y}{\partial t} \hat{j} + \frac{\partial M_z}{\partial t} \hat{k} \), the \( x \), \( y \), and \( z \) components of \( \frac{\partial \mathbf{M}}{\partial t} \) are expressed as

\[
\begin{align*}
\frac{\partial M_x}{\partial t} &= \gamma M_y (B_0 + G_z) - \frac{M_x}{T_2} + D \nabla^2 M_x - \mathbf{v} \cdot \nabla M_x, \\
\frac{\partial M_y}{\partial t} &= -\gamma M_x (B_0 + G_z) - \frac{M_y}{T_2} + D \nabla^2 M_y - \mathbf{v} \cdot \nabla M_y, \\
\frac{\partial M_z}{\partial t} &= -\frac{(M_z - M_0)}{T_1} + D \nabla^2 M_z - \mathbf{v} \cdot \nabla M_z.
\end{align*}
\]  

(B.1)
Differentiating the transverse magnetisation, \( m = M_x + iM_y \), with respect to time and substituting Equation B.1 gives

\[
\frac{\partial m}{\partial t} = \frac{\partial M_x}{\partial t} + i \frac{\partial M_y}{\partial t},
\]

\[
= \gamma M_y (B_0 + Gz) - \frac{M_x}{T_2} + D \nabla^2 M_x - \mathbf{v} \cdot \nabla M_x,
\]

\[
+ i(-\gamma M_x (B_0 + Gz) - \frac{M_y}{T_2} + D \nabla^2 M_y - \mathbf{v} \cdot \nabla M_y),
\]

\[
= -i\gamma (B_0 + Gz)m - \frac{m}{T_2} + D \nabla^2 m - \mathbf{v} \cdot \nabla m,
\]

\[
= -i\omega_0 m - i\gamma (Gz)m - \frac{m}{T_2} + D \nabla^2 m - \mathbf{v} \cdot \nabla m,
\]

(B.2)

which is Equation 4.11 in Section 4.2.2.

### B.2 IVIM model testing

Spin magnetisation was simulated on a line defined by coordinate \( \bar{x} = [0, 10 \text{ mm}] \) oriented at an angle of \( \theta = \pi/3 \) rad to the \( z \)-axis. Different types of fluid movement along the line were implemented to verify the solution accuracy of the model. \( \psi \) was fixed with a magnitude \( A = 1 \) and a phase \( \varphi = 0 \) at \( \bar{x} = 0 \) mm, and \( \psi \) was initialised to \( A(\bar{x},0) = 1 \) and \( \varphi(\bar{x},0) = 0 \). Simulations in this section were carried out with the PGSE parameters listed in Table 4.3, except for the amplitude of the gradient pulses (\( G \)) which was set to \( 1.21 \times 10^{-4} \text{ Tmm}^{-1} \).

#### B.2.1 Model accuracy

Movement of the fluid along the line \( \bar{x} \) was simulated as (1) diffusion (with \( D=2.5\times10^{-3} \text{ mm}^2/\text{s} \)), (2) uniform flow at a velocity of \( 70.7 \text{ mm/s} \), and (3) a combination of diffusion and uniform flow. The solution for \( \psi \) along \( \bar{x} \) for the different flow conditions are shown in Figure B.1.

For Case 1 (diffusion only) shown in Figure B.1a, \( A \) attenuates to a magnitude of \( 7.39\times10^{-1} \) rather than reverting to its initial magnitude of 1, due to the inability of spins to revert to their original alignment. This corresponds with the analytical
solution given by Stejskal [198]

\[ A = \exp \left( -\gamma^2 G^2 \cos^2 \theta \delta^2 \left( \Delta - \frac{\delta}{3} \right) D \right). \]  

Due to the random phase shifts caused by atomic diffusion, the average phase shifts occurring in the whole ensemble of spins within a voxel gives rise to no net shift in phase, which is also captured by the model solution for \( \varphi = 0 \) along \( \bar{x} \).

For Case 2 (flow without diffusion), the outflowing spins are replaced by inflowing spins along the direction of flow. Since the spins remain in phase, the magnitude of \( \psi \) remains unchanged as shown in Figure B.1b where \( A = 1 \) throughout \( \bar{x} \). In theory, the spins experience a change in phase which has been expressed by Stejskal [198] as

\[ \varphi = \gamma \delta G \cos \theta \Delta v. \]  

Beyond \( \bar{x} = 8 \) mm (which represents the spins that have advected from \( \bar{x} = 0 \) mm over the span of the PGSE sequence), the model solution for \( \varphi \) stabilises to 201.08 radian and is consistent to the analytical solution for \( \varphi \).

For Case 3 (diffusion and flow) as shown in Figure B.1c, spins at the inlet are advected to beyond 8 mm of the line during the course of the PGSE sequence. Spins beyond 8 mm experience signal attenuation (\( A = 7.39 \times 10^{-1} \)) due to the incoherent orientation of spins induced by diffusion. The diffusion-induced phase distribution also received a net phase shift due to the coherent flow, resulting in a \( \varphi \) of 201.08 radian. Overall, solutions generated by the model under different flow conditions correspond with analytical solutions, and demonstrate the accuracy of the model.

### B.2.2 Selection of boundary conditions

Whether the spins that enter into the solution domain are magnetised (\( A = 1 \)), or not (\( A = 0 \)), or are somewhere in between (\( 0 < A < 1 \)), depends on the distance travelled by the spin during the time-span of the PGSE sequence, which is determined by the timescales of spin movement in relation to the timescale of imaging. Here, we assess the model solution when different boundary conditions of \( A = 1 \) and \( A = 0 \) are implemented at the inlet of the domain. In the previous section, we simulated magnetisation under diffusion...
Figure B.1: Model solution for $\psi$ under different types of fluid movement. Analytical solution for the magnitude of $\psi$ ($A$) and phase ($\varphi$) are shown in red. (a) Diffusion causes random phase shifts and the incoherence of spins give rise to attenuation of magnitude of $\psi$ ($A = 7.39 \times 10^{-1}$) and no net phase shift ($\varphi = 0$). (b) Under coherent flow, spins are advected along the line to beyond $\bar{x} = 8$ mm during the span of the PGSE sequence but remain coherent ($A = 1$) while experiencing a phase shift ($\varphi = 201.08$ radian). (c) With both diffusion and coherent flow, spins advected beyond $\bar{x} = 8$ mm during the span of the PGSE sequence experience an attenuation of $A$ ($A = 7.39 \times 10^{-1}$) and a phase shift ($\varphi = 201.08$ radian).
and flow using a fixed boundary condition of $A = 1$ at the inlet and the model solution (Figure B.1c) corresponds to the analytical solution expected for such flow profile. When the boundary condition was changed to $A = 0$ for the same model geometry and flow conditions, the model solution (Figure B.2) approaches but does not reach the analytical solution ($A = 7.39 \times 10^{-1}$ and $\varphi = 201.08$), with $A$ falling short at $7.36 \times 10^{-1}$ and $\varphi$ being $201.23$ at $\bar{x} = 10$ mm. Given that a more accurate model solution was generated using an inlet boundary condition of $A = 1$, we chose to prescribe $A = 1$ at the inlet nodes for all IVIM simulations. This implies that the applied magnetic field was assumed to extend over a region larger than the solution domain such that all spins entering into the solution domain during the PGSE sequence are magnetised.

### B.2.3 Solution convergence

Convergence analysis was performed using the same geometry with diffusion and flow. The line was repeatedly refined from 50 to 1000 equally sized elements. Figure B.3a, which shows the solution for $\psi$ near the end of the line domain ($\bar{x} = 9$ mm), demonstrates that spatial convergence is attained when the number of elements was more than 100 (i.e. element size smaller than 0.1 mm). The size of timestep was varied from $1 \times 10^{-5}$ s to $8 \times 10^{-4}$ s and Figure B.3b demonstrates that convergence is attained when the size of timestep is smaller or equal to $1 \times 10^{-4}$ s.
Appendix B. IVIM model derivation and verification

Figure B.3: Convergence analysis for IVIM model. Analytical solution for the magnitude of $\psi$ ($A$) and phase ($\phi$) are shown in red. Solution for $\psi(\bar{x} = 9 \text{ mm})$ attain convergence when (a) the number of elements is more than 100 (element size $\leq 0.1 \text{ mm}$), and (b) timestep ($dt$) $\leq 1 \times 10^{-4} \text{ s}$.

B.3 Biexponential function fitting of signal attenuation curve

The signal attenuation curves generated by the IVIM model were fitted to a biexponential function (Equation 4.17). $D$, $f$ and $D^*$ were initialised with estimates. The first approximation of $D$ was determined from an exponential fit of the signal attenuation curve for $b$ greater than $10 \text{ s/mm}^2$. $D^*$ was estimated as 10 times bigger than $D$. Since $D^*$ is considerably larger than $D$, the term containing $D^*$ is negligible at high $b$ values, thereby reducing Equation 4.17 to

$$S = S(\text{high}) e^{-bD}, \quad (B.5)$$
where \( S(\text{high}) \) is the y-intercept obtained from the exponential fit of the signal attenuation curve at \( b \) greater than 10 s/mm\(^2\).

Signal attenuation at \( b \) lower than 10 s/mm\(^2\) was then fitted using the following exponential function:

\[
S = S(0)e^{-bk}, \quad (B.6)
\]

where \( k \) is an unconstrained variable. A first approximation of \( f \) was calculated from

\[
f = \frac{(S(0) - S(\text{high}))}{S(0)}. \quad (B.7)
\]

Using these estimates, \( f \), \( D \) and \( D^* \) were subsequently adjusted to fit the biexponential function using a least-squares package in Matlab.
Appendix C

Lagrange-Galerkin finite element method

We present the mathematical formulation of the Lagrange-Galerkin finite element method [208] and its application for solving Equation 4.13 and Equation 5.1, both in the form of the classical advection-diffusion-reaction equation

\[
\frac{\partial C}{\partial t} = D_C \nabla^2 C - \mathbf{v} \cdot \nabla C + f(C), \quad (C.1)
\]

where \( C \) is the unknown variable which can be the amplitude of spin magnetisation (\( \psi \)) or the concentration of oxygen in maternal blood (\( C_m \)), \( D_C \) is the diffusion coefficient of \( C \), \( \mathbf{v} \) is the velocity field, and \( f(C) \) is the source or sink term which is a function of \( C \).

Solving Equation C.1 using conventional Eulerian methods characteristically introduces oscillations and numerical diffusion in the solution near steep solution gradients if the advection term is dominant [208]. Even though accurate solutions for such advection-dominated problem can be obtained by representing the steep solution gradients using refined finite element or finite difference meshes and/or small time-steps, the computational costs are high and it is impractical for solving problems with a large mesh. For computational tractability, the Lagrange-Galerkin method, as illustrated here, has been introduced to solve advection-dominated problems.
C.1 Finite element formulation

In the Lagrange-Galerkin method, the time derivative of \( C \) is given by the Lagrangian derivative where the flow is tracked from the reference frame of a particle

\[
\frac{DC}{Dt} = \frac{\partial C}{\partial t} + v \cdot \nabla C.
\]  

(C.2)

This pure advection equation was solved for an auxiliary unknown \( C^* \) by tracing the departure point of blood at each mesh node and assigning \( C \) at the blood’s departure point to the mesh node at the start of each timestep.

Substitution of the Lagrangian derivative into Equation C.1 gives

\[
\frac{DC}{Dt} = Dc \nabla^2 C + f(C).
\]  

(C.3)

By discretising the Lagrangian derivative using explicit finite difference approximation and applying the Galerkin finite element method to the diffusion-reaction equation, the Lagrange-Galerkin formulation of Equation C.1 was obtained

\[
\int_\Omega \omega \frac{C^{n+1} - C^*}{\Delta t} = \int_\Omega \omega (Dc \nabla^2 C^* + f(C^*)) d\Omega,
\]  

(C.4)

where \( \omega \) is a weighting function and \( \Omega \) is the solution space. As \( C^{n+1} = C^* + \Delta C \), where \( \Delta C \) is the change in \( C \) in a timestep resulting from diffusion and source or sink terms, the weak formulation was obtained by applying the Green-Gauss theorem

\[
\int_\Omega \omega \frac{\Delta C}{\Delta t} = - \int_\Omega (Dc \nabla C^* \cdot \nabla \omega + f(C^*) \cdot \omega) d\Omega + \int_{\partial \Omega} Dc \nabla C^* \cdot n \, \omega d\Omega,
\]  

(C.5)

where \( n \) is the outward facing normal to the boundary and hence, the boundary condition was prescribed in the form of diffusive flux:

\[
\int_{\partial \Omega} -Dc \nabla C^* \cdot n \, \omega d\Omega.
\]  

(C.6)
C.2 Particle tracking

To determine the auxiliary unknown \( C^* \), particles located on each mesh node was traced back along its path of movement during the span of a timestep to determine where it was at the beginning of that timestep. Using the Runge-Kutta method, the departure location of each particle \( (z^*) \) at the beginning of a timestep (e.g. \( t = 0 \)) was determined from

\[
z^* = z - \int_0^{dt} v dt,
\]

(C.7)

where \( z \) is the location of the particle after a timestep \( (t = dt) \) and \( v \) is the velocity of the particle. By applying linear interpolation method, \( C \) at each of the departure point was determined using the location of the departure point. \( C \) at the departure points were assigned to \( C^* \) for the respective mesh node to signify the movement of \( C \) to the new location through advection.

For Equation 4.13 where \( \psi \) is a complex number with a magnitude \( A \) and phase \( \varphi \), \( A \) was ‘advected’ in its entirety from the departure locations to the respective mesh nodes over the span of a timestep. However, as gradient pulses are applied along the \( z \)-axis, the spin particle experiences a relative phase shift (\( \varphi_r \)) as it moves from its departure position to the mesh node:

\[
\varphi_r = \gamma \int_0^{dt} G(\zeta) \cdot z(\zeta) d\zeta,
\]

(C.8)

By applying the Euler method and trapezium rule, phase accumulated by the spins as they move to the mesh nodes over each timestep is

\[
\varphi^*(z, t_{k+1}) = \varphi(z^*, t_k) + dt \gamma G \cdot (z + z^*)/2,
\]

(C.9)

where \( \varphi^* \) is the phase at location \( z \) at the end of a timestep \( t_{k+1} \), \( \varphi(z^*, t_k) \) is the phase at the departure point at the beginning of a timestep \( t_k \).
Appendix C. Lagrange-Galerkin finite element method

C.3 Approximation space

The solution space was approximated with a linear basis function,

\[ C = \sum_{i=1}^{n\text{node}} C_i \xi_i, \quad \text{(C.10)} \]

where \( n\text{node} \) is the total number of nodes, \( i \) is the node number, \( C_i \) is the value of \( C \) at node \( i \), and \( \xi_i \) refers to the basis functions associated with node \( i \). For a 1D element with node numbering as illustrated in Figure C.1a, the linear basis function \( (\xi_i) \) associated with node \( i \) is expressed as

\[ \xi_1 = \frac{1}{\Delta x} (1 - x), \quad \xi_2 = \frac{1}{\Delta x} x, \quad \text{(C.11)} \]

where \( \Delta x \) is the length of the element.

For a 2D element with node numbering as shown in Figure C.1b, \( \xi_i \) is

\[ \begin{align*} 
\xi_1 &= \frac{1}{\Delta x \Delta y} (1 - x)(1 - y), \\
\xi_2 &= \frac{1}{\Delta x \Delta y} x(1 - y), \\
\xi_3 &= \frac{1}{\Delta x \Delta y} (1 - x)y, \\
\xi_4 &= \frac{1}{\Delta x \Delta y} xy, \\
\xi_5 &= \frac{1}{\Delta x \Delta y \Delta z} (1 - x)(1 - y)(1 - z), \\
\xi_6 &= \frac{1}{\Delta x \Delta y \Delta z} x(1 - y)(1 - z), \\
\xi_7 &= \frac{1}{\Delta x \Delta y \Delta z} (1 - x)(1 - y)z, \\
\xi_8 &= \frac{1}{\Delta x \Delta y \Delta z} x(1 - y)z, \\
\end{align*} \quad \text{(C.12)} \]

where \( \Delta x \) and \( \Delta y \) are the length of the element in the \( x \)- and \( y \)-directions, respectively.

For a 3D element with node numbering as shown in Figure C.1c, \( \xi_i \) is

\[ \begin{align*} 
\xi_1 &= \frac{1}{\Delta x \Delta y \Delta z} (1 - x)(1 - y)(1 - z), \\
\xi_2 &= \frac{1}{\Delta x \Delta y \Delta z} x(1 - y)(1 - z), \\
\xi_3 &= \frac{1}{\Delta x \Delta y \Delta z} (1 - x)y(1 - z), \\
\xi_4 &= \frac{1}{\Delta x \Delta y \Delta z} xy(1 - z), \\
\xi_5 &= \frac{1}{\Delta x \Delta y \Delta z} (1 - x)y z, \\
\xi_6 &= \frac{1}{\Delta x \Delta y \Delta z} x(1 - y)z, \\
\xi_7 &= \frac{1}{\Delta x \Delta y \Delta z} (1 - x)y z, \\
\xi_8 &= \frac{1}{\Delta x \Delta y \Delta z} xy z, \\
\end{align*} \quad \text{(C.13)} \]

where \( \Delta x \), \( \Delta y \), and \( \Delta z \) are the length of the element in the \( x \)-, \( y \)-, and \( z \)-directions, respectively.
C.4 Discretization of equation

In the Galerkin finite element method, the weighting function $\omega$ is equivalent to the basis function (i.e. $\omega = \xi_j$). Substitution of Equation C.10 in Equation C.5 gives

\[
\sum_{i=1}^{\text{node}} \frac{\Delta C_i}{\Delta t} \int_\Omega \xi_i \xi_j d\Omega = - \sum_{i=1}^{\text{node}} C_i^* \int_\Omega (D_C \nabla \xi_i \cdot \nabla \xi_j d\Omega + \sum_{i=1}^{\text{node}} C_i^* \int_\Omega f(\xi_i) \cdot \xi_j d\Omega + \int_{\partial \Omega} D_C \nabla C^* \cdot n \xi_j d\Omega.
\]  
\tag{C.14}
\]

For each element in the solution space, Equation C.14 is re-written as

\[
\sum_{k=1}^{\text{nn}} \frac{\Delta C_k}{\Delta t} \int_\Omega \xi_k \xi_j d\Omega = - \sum_{k=1}^{\text{nn}} C_k^* \int_\Omega (D_C \nabla \xi_k \cdot \nabla \xi_j d\Omega + \sum_{k=1}^{\text{nn}} C_k^* \int_\Omega f(\xi_k) \cdot \xi_j d\Omega + \int_{\partial \Omega} D_C \nabla C^* \cdot n \xi_j d\Omega,
\]  
\tag{C.15}
\]

where $\text{nn}$ is the number of nodes in an element (2 in 1D, 4 in 2D, and 8 in 3D), and $k$ is the local numbering for nodes in an element.

C.5 Assembly of equations and boundary conditions

Equation C.15 for each element in the solution space was assembled into a system of equations given by

\[
M_M \Delta C = M_K \Delta t C^* + M_F \Delta t,
\]  
\tag{C.16}
\]
where

\[ M_M = \int_{\Omega} \xi_i \xi_j d\Omega, \]
\[ M_K = \int_{\Omega} (D_C \nabla \xi_i \cdot \nabla \xi_j) d\Omega + \int_{\Omega} f(\xi_i) \cdot \xi_j d\Omega, \quad \text{and} \]
\[ M_F = \int_{\partial \Omega} D_C \nabla C^* \cdot \mathbf{n} \, \xi_j d\Omega. \]

Prescribed flux conditions (natural boundary conditions) were incorporated into \( M_F \) during assembly of the equations, while predefined \( C \) at specific node (essential boundary conditions) were imposed by replacing \( \Delta C \) as 0 at those nodes before solving the system of equations. Equation C.16 was solved for \( \Delta C \), which was added to \( C^* \) at the end of every timestep to obtain the distribution of \( C \).
Appendix D

Determining oxygen content of blood

Oxygen entering into the IVS is bound to haemoglobin or dissolved in plasma of the maternal blood. For maternal blood with an oxygen partial pressure of $pO_2$ (in mmHg)$^1$, the concentration of oxygen dissolved in the maternal plasma ($C_{\text{plasma}}$) was obtained by:

$$C_{\text{plasma}} = 3 \times 10^{-5} \times pO_2 \text{ ml/ml.} \quad (D.1)$$

For oxygen bound to haemoglobin in the maternal blood, oxygen content carried by blood is related to the partial pressure of oxygen by the haemoglobin dissociation curve. A modified Hill equation is often used to establish the amount of haemoglobin saturation in blood ($S_{Hb}$):

$$\log pO_2 = k_1 - k_2(pH - 7.4) + k_3\log(S_{Hb}/(100 - S_{Hb})), \quad (D.2)$$

where $k_1 = 1.445$, $k_2 = 0.456$, and $k_3 = 0.371$. These constants are obtained by fitting Equation D.2 to the dissociation curve derived by the mathematical model proposed by Dash and Bassingthwaighte [241].

The oxygen content carried by haemoglobin ($C_{Hb}$) is calculated from $S_{Hb}$, and the oxygen capacity of haemoglobin in the blood. Oxygen capacity is given as the product of the amount of oxygen that can be carried by haemoglobin (1.34 ml/g) and the amount

$^{1}$1Pa = $7.50 \times 10^{-3}$ mmHg
Oxygen content of blood (0.125 g/ml):

\[ C_{Hb} = \frac{S_{Hb} \times \text{O}_2 \text{ capacity}}{100}. \]  

The total concentration of oxygen carried in maternal blood, \( C_m \) (in ml/ml), is given by the sum of oxygen dissolved in the plasma and oxygen bound to haemoglobin.

\(^2\)Under atmospheric pressure \((1.01 \times 10^5 \text{ Pa})\) and body temperature \((310K)\), 1 ml/ml of oxygen in blood is equivalent to 39.3 mol/m³.
Appendix E

Verification of oxygen transport model

Here we perform verification of the oxygen transport model by parameterising the model with parameters from Chernyavsky et al. [55] and comparing our model’s results with their results.

E.1 Model parameterisation and output metrics

Table E.1 lists the parameter values used in the oxygen transport model and Chernyavsky et al. [55]. We followed most of the parameters used in Chernyavsky et al. [55]. The main difference lies in the choice of domain geometry. Unlike the 3D hemisphere implemented in the model of Chernyavsky et al. [55], we represented the placental subunit as a 2D rectangle. We adopted the diameter and radius of Chernyavsky et al.’s 3D hemisphere as our domain width and thickness respectively. As the model domain used by Chernyavsky et al. [55] is essentially a 3D axisymmetric medium, we do not anticipate the differences in model geometries will significantly influence model predictions. In view of the different model geometry, we fitted the placental hydraulic conductivity ($\kappa$) and the oxygen uptake constant ($\alpha$) so that the relative and absolute net solute uptake rate for the baseline case correspond with the rates generated by the model of Chernyavsky et al. [55]. As the solute concentration fields were all normalised with a reference concentration ($C_0$)
Appendix E. Verification of oxygen transport model

Table E.1: Model parameterisation based on Chernyavsky et al. [55]. Parameters were chosen to verify our model against the model of Chernyavsky et al. [55].

<table>
<thead>
<tr>
<th>Symbol</th>
<th>Description</th>
<th>Model value</th>
<th>Model values in Chernyavsky et al. [55]</th>
</tr>
</thead>
<tbody>
<tr>
<td>$\tau$</td>
<td>Domain thickness</td>
<td>20 mm</td>
<td>20 mm</td>
</tr>
<tr>
<td>$\epsilon$</td>
<td>Domain width</td>
<td>40 mm</td>
<td>40 mm</td>
</tr>
<tr>
<td>$x_v$</td>
<td>Distance between SA and DV</td>
<td>18 mm</td>
<td>18 mm</td>
</tr>
<tr>
<td>$d$</td>
<td>Diameter of SA and DVs</td>
<td>2 mm</td>
<td>2 mm</td>
</tr>
<tr>
<td>$d_{TV}$</td>
<td>Terminal villi diameter</td>
<td>0.05 mm</td>
<td>0.05 mm</td>
</tr>
</tbody>
</table>

**Maternal blood flow model**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Description</th>
<th>Model value</th>
<th>Model values in Chernyavsky et al. [55]</th>
</tr>
</thead>
<tbody>
<tr>
<td>$\mu$</td>
<td>Blood viscosity</td>
<td>$4 \times 10^{-3}$ Pa.s</td>
<td>$4 \times 10^{-3}$ Pa.s</td>
</tr>
<tr>
<td>$q_0$</td>
<td>Reference inlet blood flow</td>
<td>$83.3$ mm$^3$/s (5 ml/min)</td>
<td>$83.3$ mm$^3$/s (5 ml/min)</td>
</tr>
<tr>
<td>$\kappa$</td>
<td>Hydraulic conductivity in IVS</td>
<td>$4.5 \times 10^{-4}$ mm$^2$</td>
<td>$\sim 10^{-4}$ mm$^2$</td>
</tr>
</tbody>
</table>

**Oxygen transport model**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Description</th>
<th>Model value</th>
<th>Model values in Chernyavsky et al. [55]</th>
</tr>
</thead>
<tbody>
<tr>
<td>$D_{O_2}$</td>
<td>Diffusivity of oxygen in blood</td>
<td>$1.62 \times 10^{-3}$ mm$^2$/s</td>
<td>$1.67 \times 10^{-3}$ mm$^2$/s</td>
</tr>
<tr>
<td>$\alpha_0$</td>
<td>Reference oxygen uptake constant</td>
<td>$0.125$ s$^{-1}$</td>
<td>$0.0167$ s$^{-1}$</td>
</tr>
<tr>
<td>$C_0$</td>
<td>Oxygen concentration entering through SA</td>
<td>$6.44$ mol/m$^3$ (0.164 ml/ml)</td>
<td>$0.1$ mol/m$^3$</td>
</tr>
</tbody>
</table>

in Chernyavsky et al’s model, we chose to use the oxygen concentration estimated at the SA inlet as our reference solute concentration [226].

Following Chernyavsky et al. [55], we applied a maternal blood flow flux $Q_m$ into the placentone as a boundary condition. In Chernyavsky et al. [55], the transport and uptake problem have been characterised by an uptake parameter $Da$ (also known as the Damköhler number) and a geometrical ratio $h_{DV}$. $Da$ represents the local solute consumption rate by the villous tissue relative to the rate of mass transfer by maternal blood and is defined as $Da = \alpha L^3 / Q_m$, where $L$ is the radius of the placentone (i.e. half of the domain width for our model), while $h_{DV}$ represents the position of DVs relative to the placentone boundary, and is expressed as $h_{DV} = x_v / L$. The outputs of Chernyavsky et al’s model are given by the absolute net uptake rate ($N_a / q_0 C_0$) and net relative uptake rate ($N_T$). $N_a$ is the difference between the concentration flux at the SA ($Q_m C_0$) and the concentration fluxes at the DVs. The relative net uptake rate is defined as $N_a / Q_m C_0$, which is a measure of solute consumption relative to the available flux of solute. For purpose of comparison, the same dimensionless parameters and output metrics were adopted for this verification study.
E.2 Model results

The results generated by Chernyavsky et al.’s model are not included in this Appendix but can be found in [55]. Using parameters listed in Table E.1, Figure E.1a shows the flow streamlines generated by our model for the baseline case ($Da = 1$ and $h_{DV} = 0.9$). The flow streamlines follow the same pattern as that generated by Chernyavsky et al. [55], with the streamlines starting from the SA inlet extending towards the boundary of the domain and terminating at the DVs. The pressure difference drop across the placental subunit (approximately 800 Pa (6 mmHg)) is also close to the value reported by Chernyavsky et al. [55]. Oxygen distribution for the baseline case (Figure E.1b) also exhibits the same pattern as predicted by Chernyavsky et al. [55]. When the distance between the SA and DV was reduced ($Da = 1$ and $h_{DV} = 0.45$) and the uptake parameters were varied (low uptake where $Da = 0.25$ and $h_{DV} = 0.9$, and high uptake where $Da = 4$ and $h_{DV} = 0.9$), Figures E.1c–f show that the model is able to reproduce the same flow and solute transport behaviour under these scenarios. Like in the model of Chernyavsky et al. [55], the pressure drop in the IVS (667 Pa (5 mmHg)) is also lower as compared to the baseline case when the distance between the SA and DV was reduced ($Da = 1$ and $h_{DV} = 0.45$). Table E.2 shows the relative and absolute net uptake rate generated by the model for different $Da$ and $h_{DV}$. All estimates are close to the values predicted by Chernyavsky et al. [55].

Figure E.2 illustrates that our model exhibits similar behaviour as predicted by Chernyavsky et al. [55] when different model parameters were varied while keeping the other parameters constant. Figure E.2a demonstrates that oxygen distribution and uptake improve when the DVs were located nearer to the peripheral of the placentone boundary. Oxygen uptake increases with the uptake constant ($\alpha$) (Figure E.2b). Figure E.2c shows that while the absolute net uptake rate increases with higher blood flow rate $Q_m$, the relative net uptake rate decreases. As predicted by Chernyavsky et al. [55], our model also exhibits an optimal uptake rate at a villous tissue volume fraction ($\phi$) of 0.25. Based on the presented results, we demonstrated that our model is able to replicate the results of Chernyavsky et al. [55] using our current methodology, and hence, have verified the accuracy of our model.
Figure E.1: Results generated by model parameterised based on Chernyavsky et al. [55]. (a) Streamlines (blue lines) and (b) normalised concentration field (colours) generated for the baseline case \((D_a = 1\) and \(h_{DV} = 0.9\)). (c) Streamlines and (d) normalised concentration field when the distance between the SA and DV was reduced \((D_a = 1\) and \(h_{DV} = 0.45\)). Normalised concentration fields when the uptake parameter is (e) low \((D_a = 0.25\) and \(h_{DV} = 0.9\)), and (f) high \((D_a = 4\) and \(h_{DV} = 0.9\)). Oxygen concentration is normalised by oxygen concentration of maternal blood entering the IVS.
E.2. Model results

Table E.2: Uptake rate predicted by model parameterised based on Chernyavsky et al. [55]. Relative net uptake rate ($N_T$) and absolute net uptake rate ($N_a/q_0C_0$) predicted by the model with different relative distances between the SA and DV ($h_{DV}$) and different uptake parameters ($Da$).

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Figure E.2: The effects of placental structure on oxygen uptake predicted by model parameterised based on Chernyavsky et al. [55]. The model predicts the same behaviour in oxygen uptake when (a) the relative distance between the SA and DV to the width of the placentone ($h_{DV}$), (b) uptake constant ($\alpha$), (c) blood flow rate at the SA ($Q_{in}$), and (d) villous tissue volume fraction ($\phi$), were varied.


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