Particle separation in low Re flow conditions:
How do red blood cells decide which branch to take?

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Overview

- The microcirculation
  - Oxygen and nutrient delivery to the cells
- Blood: Newtonian fluid?
  - 2-phases: The plasma and the cells
  - Fahraeus Effect: Cells travel in the middle
  - Plasma skimming: Side branches get more plasma
  - Low Reynold’s flow: Viscous nature predominates
  - Bifurcation shapes in vivo: RBC filters
- Large Scale Model
- Microchannel Model
Systemic vs. microcirculatory systems

- **Systemic circulation**
  - >200μm dia.
    - Organ inflow vessels
  - **Pressure**
    - 120/80 mmHg to 5 mmHg
  - **Velocity (aorta)**
    - 800 cm/min
  - **Shear stress**
    - 20 - 50 dyn/cm²
  - **Re >> 1**
    - Inertia predominates
Systemic vs. microcirculatory systems

- **Microcirculation**
  - < 200µm dia.
    - particle to tube dia. ratio
  - Static pressure
    - 50 - 0 mmHg
  - Fluid velocity
    - 0 - 6000µm/sec
  - Fluid shear stress
    - 0 - 80 dynes/cm²
  - Re << 1 (Reynolds number)
    - Viscous resistance predominates

neuro.wehealny.org
Human Hair Thickness

- 50 - 200 microns in diameter
Lab-on-a-chip?

- The principles learned from our studies apply to any microchannel system with particles.
  - Blood analysis chips
  - Bio-hazard detection
- The relative sizes of the tube (channel) and particles are what is important.
Newtonian fluids and the continuum concept

- Newtonian fluids are defined as continuous.
  - Continuum fluid – all of the pieces of the fluid have the same composition and nature
  - Continuum flow – all of the moving streamlines are uniform and seamless

- How small can a volume of blood be and still appear to be continuous?
Blood is a 2-phase fluid: plasma phase and particle phase

- **Hematocrit**
  - Red Blood Cells
    - 5µm
    - 30 - 40%
- **White Blood Cells**
  - 7 - 10µm
- **Platelets**
  - 1 - 3µm
- **Plasma proteins**
  - 20 - 80nm

http://www.nature.com/nm/journal/v9/n5/images/nm0503-481-I1.jpg
A note about the ‘tube’ – the vascular wall

■ Endothelial cells form the inside lining.
  □ Must withstand fluid shearing forces

■ Vascular smooth muscle cells wrap outside.
  □ Actively constrict and dilate to change vessel diameter
Tube to particle diameter ratio

Ratio 1:1
- Capillaries
- Slug flow
- RBC tube

Ratio 1:2
- Small arterioles
- No theory to accurately describe the ‘flow properties.’

Ratio 1:20
- 50 - 200µm
- Bubble flow
- Newtonian fluid with Newtonian flow properties

Systemic circulation

Microcirculation
In vivo microvascular networks

Frame Lab: endothelial cells stained with BS-1 lectin

Brightfield and epifluorescence

XRITC labeled erythrocyte

MVR, 1993
Non-Newtonian 2-phase flow

- Particles and plasma move differently from each other
Non-Newtonian 2-phase flow

- Velocity profile
  - Faster flow in the centerline
- RBC travel in faster centerline
  - Cell free zone near the wall
  - RBC move faster than the whole blood
Fahraeus Effect

- RBC move faster than the plasma because they are only in the fast streamlines.
Plasma skimming

- At a branch point, if 30% of the flow (volume = nl/min) travels to the branch,
- less than 30% of the RBC (flux = #RBC/time) travel to the branch.

[Diagram of blood flow showing capillaries feeding downstream and branches with oxygen distribution indicated]
How do the RBCs decide which branch to take?

- Plasma skimming is common knowledge.
- There are no models to accurately predict the degree of skimming that will occur.
- Particle flux (oxygen) distribution is therefore not predictable.
- This severely restricts our ability to understand and treat peripheral vascular disease or damage.
Translational Focus 1: Diabetes

Diabetics have fewer microvessels to begin with.

Non-uniform oxygen delivery means:

- Decreased ability to keep up with oxygen demand during exercise
- Decreased ability to increase oxygen delivery to heal wounds
Translational Focus 2: Wound healing

Anthony Dewar, MS, December 2009!!!

- The key rate limiting step in healing wounds is delivery of enough oxygen to keep up with the increased metabolism.

- Both flow through existing microvessels and creation of new microvessels (angiogenesis) are crucial.
To understand oxygen delivery we must understand 2-phase particle flow in the microcirculation.

- Particle motion within the tube ✓
  - Consequence for particle flow path
- Inertial forces relative to viscous resistance
  - Consequence for peak shearing forces
- Shape of the branching region (bifurcation)
  - Consequence for particle exclusion
Reynold’s number

- Ratio of inertial forces to viscous resistance

\[ \text{Re} = \frac{\rho \bar{u} D}{\eta} \]

- \( \rho \), density, gm/cm \( \rightarrow \) assumed constant
- \( \bar{u} \), mean axial fluid velocity, cm/s \( \rightarrow \) shear rate
- \( D \), tube diameter, cm
- \( \eta \), viscosity, gm/cm*\( s \)

A note about multidisciplinary research: Mechanical Engineers use \( \mu \) to denote viscosity. For a Chemical Engineer, \( \mu \) is electrochemical potential; \( \eta \) is viscosity.
Re - perspective

- Water from a fire hose
  - 110
- Blood from the aorta
  - 3500 → inertia dominates
- Blood in a typical capillary
  - 0.1 to 0.01 → viscosity dominates
Inertial forces: shear rate

- **Shear rate, s\(^{-1}\)**
  - \(\frac{dv}{dr}\), initial slope of velocity profile

- **Shear stress, dyn/cm\(^2\)**
  - \(\tau_\omega = \eta \times \frac{dv}{dr}\)
  
  Shear stress = viscosity * shear rate

Does the shearing force predict RBC distribution at branches?
For low Re, peak shear is predicted to occur before the branch opening.

Branching angle, $\phi$, range from 30 - 150°

Computational fluid dynamics model

Biorheology, 37: 325-340, 2000
In vivo data: peak shear before branch.

Shear relative to inflow

Circumference ~ 50 µm

4 cells form lining

Endothelial cells see high shear

Endothelial cells see low shear
Hypothesis:

- Bifurcations with highest peak shear on the lateral wall will have the highest RBC flux to the branch.
Peak Shear on Lateral Wall is unrelated to RBC flux.

What about the shape of the flow divider?
2-D projected shape of intersection

Biorheology, 37: 325-340, 2000
Hypothesis:

- Bifurcation shape (angle) influences RBC flux to the branch.
Bifurcation shape and flux are related differently by branch location.
Further, bifurcation angles change in vivo with uniform dilation.
Let’s control something – Large Scale Model

PDMS particle 0.5 cm diameter

Distance is Θ dependent.

Initial thesis project: Aparna Kadam, PhD candidate
Large Scale Model System

Syringe Pumps control Bulk Flow

PDMS Particles flow in glycerol

Particle Flux, $F$, counted from recorded image

Bifurcation

Schematic courtesy of Kathleen Burke
Scaling Up the Relevant Parts

- Re 0.2 to 0.01
  - Controlled by regulating velocity in the feed
- Suspending fluid – glycerol
- RBC – PDMS polymer with correct density
- Bifurcation angles – 45, 90, 135 degrees
Hypothesis:

- Particle flux (F) to the branch will match Bulk flow (Q) to the branch.

Q = 50%
F = 50%
Bulk flow (Q) vs. particle flux (F)

Q = 1.5(±0.2) x F -29(±10); R² = 0.93

Contributing Undergraduates:
Kathleen Burke, Farha Islam, Hench Wu
Large Scale Model: effect of Re

As we lower Re, this disparity is worse.
Large Scale Model: effect of angle

Next computational model: Steven Leigh, PhD candidate
Angle has a larger effect below $Re \ 0.06$

Next computational model: Steven Leigh, PhD candidate
Points to consider

- PDMS particles are more rigid than RBC
- The bifurcation shape is sharp
  - In vivo, radius of curvature is more gradual

MVR, 1993
2-D Uniform Viscosity Model
Shear Stress Distribution along the Top Wall
Re = 0.01, 10 µm Vessel Diameter

90 Degree Bifurcation Angle
Changing radius of curvature - both sides

Increasing radius of curvature decreases the maximum shear by 33%, and shifts the maximum shear upstream.

In collaboration with Risa Robinson, PhD, RIT
To-Scale Model

- Microchannels molded from PDMS
  - Prescribe radius of curvature
  - Channel shape is square

- Particles – RBCs
Chamber Mixer Design

Senior Design Project 2009: Jason Hamilton, Johanna Sisalima
Microchannel Design

- Photolithography
  - SU-8
- Preliminary data

Thesis Project: Aparna Kadam, PhD candidate
In summary…

- What factors control plasma skimming?

  - Re < 0.06 → Lower inertia
    - (Higher internal resistance – viscosity)
    - Increased angle to the branch
    - Decreased Flux to the branch

- Angle? Radius of curvature? Particle to tube diameter ratio?
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