Blood Flow: Current State and Future Prospects

9 - 11 October 2017
Institut Henri-Poincare
Paris, France
Aims and Scope

Blood plays an essential role in many physiological and pathological processes of the organism, which motivates large scientific efforts to understand its behavior at the macro-flow scale as well as at the level of single cells. Blood is a complex fluid that exhibits a number of non-Newtonian characteristics, including viscoelasticity, shear-thinning, and yield stress. These properties result from viscoelastic characteristics of blood cells and their intricate behavior in a complex flow environment. Modern in vivo and in vitro experimental techniques in combination with sophisticated computational approaches allow nowadays to describe blood flow at the macroscale and to relate it to the behavior and interactions of single blood cells.

The main aim of the conference is to discuss current state of research on blood flow and its future prospects. This interdisciplinary conference will gather experimentalists and theoreticians to discuss mechanical, physical, and biomedical aspects of blood flow. Even though the main focus is intended to be on biophysical aspects of blood cells and blood flow, practical and clinical applications related to blood flow will be also part of the conference.

The topics will include, but are not limited to:

- Blood cell mechanics
- Hemorheology
- Hemodynamics
- Blood cell behaviour in flow
- Blood cell adhesion
- Biomedical applications
- In vivo and in vitro approaches
- Blood diseases and disorders
- Model systems including vesicles and capsules
- Theory and modelling
Conference venue

The conference will take place in the Henri Poincare Institute located in Paris, France. The Henri Poincare Institute is located in the heart of the 5th arrondissement of Paris close to the Pantheon and the Jardin du Luxembourg, and is one of the oldest and most dynamic international structures dedicated to mathematics and theoretical physics.

The address of the Henri Poincare Institute is: 11 rue Pierre et Marie Curie, 75231 Paris Cedex 05 (see the map below).
Conference organizers

- Dmitry Fedosov (Forschungszentrum Jülich, Germany)
- Alexander Farutin (CNRS, Laboratoire Interdisciplinaire de Physique, France)
- Chaouqi Misbah (CNRS, Laboratoire Interdisciplinaire de Physique, France)
- Christian Wagner (Saarland University, Germany)

Funding and support

- Forschungszentrum Jülich
- Laboratoire Interdisciplinaire de Physique
- Alexander von Humboldt Foundation
- CNRS
- University of Grenoble Alpes
- Saarland University
### Scientific program

(PL - plenary talk, IT - invited talk, CT - contributed talk)

#### Monday, 9 October 2017

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<td>“Impact of red blood cell rigidity on the vascular wall adhesion of neutrophils: implication in the pathology of sickle cell disease”</td>
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<td>“Non-adsorbing macromolecules induce adhesion of diabetic red blood cells to the endothelium”</td>
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15:30 – 15:50 CT-8: Guy Cloutier
“A protocol for in vivo measurements of erythrocyte aggregation using ultrasound spectroscopy”

15:50 – 16:20 Coffee break

16:20 – 16:50 IT-4: Thomas Podgorski
“Order and disorder in red blood cell traffic”

16:50 – 17:10 CT-9: Netanel Korin
“RBC dynamics in microfluidic capillary models of organ-specific microvasculatures”

17:10 – 17:30 CT-10: Adlan Merlo
“An in vitro study of highly confined blood flows: from single bifurcations to 2D-networks”

17:30 – 17:50 CT-11: Joana Fidalgo
“Biomimetic bifurcating networks for studying RBC partitioning in microvasculature”

18:10 – 20:00 Poster session

Tuesday, 10 October 2017

9:00 – 9:40 PL-2: Timothy Secomb
“Blood flow in the microcirculation”

9:40 – 10:10 IT-5: YongKeun Park
“Holotomography techniques for 3-D label-free imaging of live cells and tissues”

10:10 – 10:30 CT-12: Viviana Claveria
“Possible self-margination of sickle red blood cells suspensions in glass capillaries”

10:30 – 10:50 CT-13: Emilie Franceschini
“Experimental ultrasound characterization of red blood cell aggregation: estimation of the aggregate size distribution”

10:50 – 11:20 Coffee break

11:20 – 11:50 IT-6: Anne-Virginie Salsac
“Motion of a spherical capsule flowing in a branched tube with finite inertia”

11:50 – 12:10 CT-14: Prosenjit Bagchi
“Direct simulation of cellular-scale blood flow in microvascular networks”

12:10 – 12:30 CT-15: Jules Dichamp
“Modelisation of blood perfusion into a whole adipose tissue vascular network”
12:30 – 12:50 CT-16: Zaiyi Shen
“Deformability-influenced delivery of red blood cells in microvascular networks”

12:50 – 14:20 Lunch

14:20 – 14:50 IT-7: Sergey Shevkoplyas
“The dynamics of blood flow in artificial microvascular networks”

14:50 – 15:10 CT-17: Lars Kaestner
“Red blood cell passage of small capillaries is associated with transient Ca(2+)-mediated adaptations”

15:10 – 15:30 CT-18: Mathilde Reyssat
“Production of blood platelets in a microfluidic chip”

15:30 – 15:50 CT-19: Annemiek Cornelissen
“Tissue growth pressure drives early blood flow in the chicken yolk sac”

15:50 – 16:20 Coffee break

16:20 – 16:50 IT-8: Jonathan Freund
“Stability of flowing red blood cell trains”

16:50 – 17:10 CT-20: Alexander Alexeev
“Mesoscale modeling of blood clot contraction”

17:10 – 17:30 CT-21: Georgy Guria
“Platelet activation in intensive blood flows”

17:30 – 17:50 CT-22: Bastien Chopard
“A physical description of platelet deposition”

17:50 – 18:10 CT-23: Britt van Rooij
“Cell-based platelet aggregation modelling in Hemocell”

20:00 Conference dinner

Wednesday, 11 October 2017

9:00 – 9:40 PL-3: Manouk Abkarian
“On the importance of red cells deformability in blood flow”

9:40 – 10:10 IT-9: Paolo Decuzzi
“Rational design of nanoconstructs for the vascular administration of therapeutic and imaging agents”

10:10 – 10:30 CT-24: Philippe Connes
“Red blood cell rheology and vascular dysfunction in sickle cell disease”
10:30 – 10:50  CT-25: Yi-Fan Wu
   “Physical investigation of hemo-rheological characteristics for cardiac surgery patients”

10:50 – 11:20  Coffee break

11:20 – 11:50  IT-10: Ming-Chih Lai
   “Vesicle dynamics and electro-hydrodynamics: modeling and computation”

11:50 – 12:10  CT-26: Petros Koumoutsakos
   “Uncertainty quantification of the red blood cell model”

12:10 – 12:30  CT-27: Benoit Pier
   “Hydrodynamics of pulsatile flows”

12:30 – 12:50  CT-28: Anil Dasanna
   “Rolling adhesion of malaria-infected red blood cells”

12:50 – 14:20  Lunch

14:20 – 14:50  IT-11: Thomas Fischer
   “On the reference configuration of the red cell membrane”

14:50 – 15:10  CT-29: Victor Steinberg
   “Intermediate regime and a phase diagram of red blood cell dynamics in a linear flow”

15:10 – 15:30  CT-30: Lionel Bureau
   “Soft biolubrication: lift forces at a vascular wall mimic”

15:30 – 15:50  CT-31: Stephan Quint
   “3D tomography of red blood cells in micro-channels”

15:50 – 16:00  Wrap-up and closing
List of posters

P-1: Revaz Chachanidze  “Margination of blood cells”
P-2: Alberto Mantegazza  “In vitro measurements of apparent intrinsic viscosity in function of tube hematocrit and red blood cell velocity”
P-3: Sylvain Losserand  “Dispersion and transit time of red blood cells in capillaries”
P-4: Mehdi Inglebert  “Blood flow in biomimetics micro-channels”
P-5: Andreas Passos  “Velocity, haematocrit and aggregation characteristics of stiffened RBC suspension flows”
P-6: Efstathios Kaliviotis  “Blood microstructure and viscosity in bifurcating microfluidic flows”
P-7: Alexander Kihm  “Shapes and positions of red blood cells in capillary flow”
P-8: Victoria Vitkova  “Rheology of concentrated red blood cell suspensions and cell dynamics in flow”
P-9: Francois Yaya  “Flow vortices induced by red blood cells”
P-10: Marianne Fenech  “Effects of red blood cell aggregation on microparticle margination in human blood”
P-11: Yannick Knapp  “A multidisciplinary/multimodal approach to study the prothrombotic effect of shear rate gradient”
P-12: Valerie Deplano  “Red blood cell ghost suspensions as a blood mimicking fluid for microfluidic experiments”
P-13: Thomas John  “Shape and position of moving red blood cells in microchannels”
P-14: Rekha Selvan  “Role of human red blood cell membrane stiffness on time of passage through narrow straight channels”
P-15: Guy Cloutier  “Influence of erythrocyte aggregation on radial migration of platelet-sized spherical particles in shear flow”
P-16: Olivera Korculanin  “Breakup of red blood cells aggregates”
P-17: Iveta Jancigova  “Torque-free elastic moduli in spring network model of red blood cell”
P-18: Christian Bächer  “Microparticle margination in complex geometries”
P-19: Benjamin Czaja  “The effects of pulsatility in 2D cell resolved blood flow simulations of curved vessels with aneurysms”
P-20: Gabor Zavodszy  “In-silico investigation of the effect of cytoplasm viscosity on blood transport mechanics”
P-21: Amy Smith  
“Structural and hemodynamic comparison of anatomical and synthetic cerebral capillary networks”

P-22: Masoud Hoore  
“Demargination and mechanical dissociation of platelet aggregates in blood stream”

P-23: Vincent Doyeux  
“Upscaling mass transfer in brain capillary networks”

P-24: Chih-Tang Liao  
“In silico investigation of rheological characteristics of a red blood cell suspension in a simple shear flow”

P-25: Alexander Farutin  
“Hydrodynamic crystals in weakly confined shear flow”

P-26: Maxime Berg  
“Red blood cells distribution in microvascular networks: a model derived from experiments”

P-27: Brenna E. Hogan  
“Effect of red blood cells on wall shear stress and flow properties using lattice Boltzmann-immersed boundary methods”

P-28: Miguel O. Bernabeu  
“A computational study on the effect of plasma skimming on vascular development”

P-29: Aymen Laadhari  
“Fully implicit finite element methodology for the modeling of biomembranes and red blood cells”

P-30: Zakaria Boujja  
“2D simulation of red blood cells in capillaries”

P-31: A. Nait Ouhra  
“Does a vesicle migrate to the center or to the periphery in a bounded shear flow?”

P-32: Myriam Peyrounette  
“Toward whole brain simulations of blood flows in human cerebral microcirculation: hybrid modelling and high performance computing”

P-33: Wei Chien  
“Different dynamics of red blood cells define their trajectory in deterministic lateral displacement arrays”

P-34: Hengdi Zhang  
“Lateral migration of vesicle in bounded Poiseuille flow: a numerical investigation on effect of viscosity contrast and initial position”

P-35: Matthias Laumann  
“Cross-stream migration of asymmetric particles driven by oscillating shear”

P-36: Winfried Schmidt  
“Migration of soft microparticles in a modulated Poiseuille flow with the flow along the grooves”

P-37: Andre Förtsch  
“Migration reversal of soft particles in vertical flows”
Plenary talk abstracts
The non-nucleated red cell is unique among human cell type in that the plasma membrane, its only structural component, accounts for all of its diverse antigenic, transport, and mechanical characteristics. Our current concept of the red cell membrane envisions it as a composite structure in which a membrane envelope composed of cholesterol and phospholipids is secured to an elastic network of skeletal proteins via transmembrane proteins. Structural and functional characterization of the many constituents of the red cell membrane, in conjunction with biophysical and physiologic studies, has led to detailed description of the way in which the remarkable mechanical properties and other important characteristics of the red cells arise, and of the manner in which they fail in disease states. A spectrin-based cytoskeleton that provides elasticity and mechanical stability necessary to survive the shear forces within the microvasculature. The architecture of this membrane skeleton and the nature of its intermolecular contacts determine the mechanical properties of red cells. The oligomeric state of spectrin is in a dynamic equilibrium that facilitates remodeling of the network as the cell changes shape in response to shear stress. Current studies in this very active and exciting field are continuing to produce new and unexpected revelations on the function of the red cell membrane and thus of the cell in health and disease, and shed new light on membrane function in other diverse cell types.
Blood flow in the microcirculation

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The microcirculation consists of an extensive network of blood vessels with diameters ranging from about 4 to 200 µm. I will review current understanding of blood flow in the microcirculation and indicate directions for future work [1]. Inertial effects are negligible and the equations of Stokes flow apply. The flow properties of blood are strongly influenced by the 40-45% hematocrit, i.e. volume fraction of red blood cells (RBCs). The typical diameter of an undeformed human RBC is about 8 µm. Consequently, blood flowing in microvessels is not well represented by continuum models. Analysis of blood flow in microcirculation requires consideration of the motion and deformation of individual RBCs. The non-continuum behavior of blood in the microcirculation leads to two physiologically important effects: strong dependence of apparent viscosity on vessel diameter (Fåhraeus-Lindqvist effect) and uneven partition of hematocrit in the downstream vessels at diverging bifurcations (phase separation effect). The formation of a cell-free or cell-depleted layer near the vessel walls is the main reason for these distinctive effects. The reduction in cell concentration in the high-shear region near the walls causes the reduction in overall apparent viscosity. The reduced hematocrit entering a low-flow branch at a bifurcation occurs because the flow entering such a branch comes preferentially from the periphery of the upstream parent vessel.

Several physical phenomena contribute to migration of RBCs away from vessel walls. (i) The finite size of the RBC limits the radial distribution of cell centers. The width of an RBC is at least 2 µm, so the center of mass of the cell cannot physically approach within 1 µm of the wall. (ii) RBCs in a shear flow adjacent to a wall tend to migrate away from the wall. (iii) The curvature of the velocity profile in tube flow produces a tendency for migration toward the centerline of the flow. (iv) Microvessels are lined with a glyocalyx or endothelial surface layer, consisting of a matrix of macromolecules, which creates an exclusion zone for flowing RBCs. An opposing tendency results from shear-induced diffusion or dispersion. In shear flow of a concentrated suspension, frequent particle-particle interactions lead to net migration down the concentration gradient, toward the walls.

Analyzing the flow of concentrated suspensions of deformable particles is a challenging problem in fluid mechanics. Migration of flexible particles across fluid streamlines involves a complex interaction between flow forces and resulting deformation and orientation of particles. Rigid particles in dilute suspension do not show net migration across streamlines in Stokes flow, so effects of deformability and particle-particle interactions are crucial. In recent years, advanced computational techniques have allowed detailed simulations of flow of large numbers of interacting particles in three dimensions, and the formation of a cell-free layer has been effectively simulated. However, a general mechanistic understanding of this phenomenon remains elusive and represents a direction for future work.

References

Blood is the archetype of a shear thinning fluid. This property common to many complex fluids seems to be essential for an efficient perfusion of the vascular tree by the heart. Shear thinning is intimately related to the dynamics and mutual interactions of RBCs, the major component of blood. Because of the lack of knowledge about the behavior of RBCs under physiological conditions of viscosity and stress, the link between RBCs dynamics and blood rheology remained unsettled for the major part of the twentieth century. I will review in this presentation the current understanding of blood shear thinning and its relationship with red blood cells local behavior. In contrast to the current paradigm, which assumes that RBCs align steadily around the flow direction while their membrane and cytoplasm circulate like a tank-tread, I will show the rich dynamics displayed by RBCs under shear flow even for semi-dilute volume fractions typical of the microcirculation. I will discuss the link between these dynamic morphologies and both in-plane elasticity of the membrane and internal viscosity of the cells. I will finally discuss the importance of these observations for physiologically relevant phenomena and how it might be important in pathological blood rheology and flow.
Invited talk abstracts
Dynamics of individual healthy, sickle and spherocytic RBCs: from very low mechanical stresses under shear flow to extreme ones in splenic-like slits

Annie Viallat

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The static and dynamic deformabilities of red blood cells (RBCs) and their kinematic behavior in microflows have been extensively studied because they are important determinants of microcirculatory blood flow [1,2]. Indeed, despite their ‘simple’ solid-liquid composite structure, RBCs under shear flow exhibit a large variety of shapes [3], can extrude tens of micron long tethers, and display many different regimes of motion (such as well-known tumbling and tanktreading). Even today, the rheology and the kinematics of the red blood cells under flow can reserve surprises. Here, we first focus on little studied RBC motions, flip-flopping and rolling, observed in shear flow at low shear stress and very small deformation. We report the evolution of the RBC orientation in the flow with the applied shear stress and the effect of RBC density and sickle cell and thalassemia diseases. We then focus on the dynamics of single RBCs submitted to extreme deformations when they pass through biomimetic splenic-like slits of submicron widths [4]. We show that RBC shapes present generic pattern at the slit exit that we discuss and compare to results of numerical simulations. We also show a novel mode of deformation of RBCs when they pass through the smallest slits with the formation of a peculiar micron-size tip at their front. We discuss the origin of the observed behavior.

Left: schematic of the splenic-like slit device. Right: RBCs with a front tip exiting a slit (5µm high, 1.9 µm long, 0.8 µm wide). Optical microscopy (x100), in-slit pressure drop 55 Pa/µm

The project has received funding from Excellence Initiative of Aix-Marseille University – A*MIDEX, a French “Investissements d’Avenir” programme.

References

Deformation and the Resultant Motion of a Red Blood Cell

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Red blood cells (RBCs) deform and change their orientations under flow. Such motions alter rheological and diffusive properties of the suspension of RBCs, thus important in understanding transport phenomena of blood. In this background, we have investigated reorientation of an RBC in shear flow [1], rheology of an RBC suspension in the dilute limit [2], and the sedimentation of an RBC under gravity [3]. The RBC was modelled as a capsule with a two-dimensional hyper-elastic membrane. Large deformation of the thin membrane was calculated by a finite element method. Due to the small size of the RBC, fluid motion around the RBC was assumed to follow Stokes flow and was solved by a boundary element method [4]. In this article, we briefly review these studies.

In [1], the motion of a spheroidal capsule or a red blood cell in creeping shear flow was investigated. The results showed that the orientation of a non-spherical capsule is variant under time reversal, though that of a rigid spheroid is invariant. Surprisingly, the alignment of a non-spherical capsule over a long time duration shows a transition depending on the shear rate.

In [2], we performed a numerical investigation of the rheological properties of an RBC suspension in the dilute regime, in which the bulk stress of the suspension was calculated by the stresslet generated on a single RBC. Interestingly, the effective shear viscosity of the dilute suspension might decrease with increasing viscosity of the internal liquid.

In [3], the reorientation phenomenon of a single red blood cell during sedimentation was investigated. The cell settles downwards due to a density difference between the internal and external fluids, and it changes orientation toward a vertical orientation regardless of Bond number or viscosity ratio. The reorientation phenomenon is explained by a shape asymmetry caused by the gravitational driving force, and the shape asymmetry increases almost linearly with the Bond number. As a result, the relaxation time of reorientation was proportional to \( g^{-2} \), where \( g \) is the gravitational acceleration.

These studies revealed that the motions of RBCs are strongly affected by the membrane deformation. Therefore, establishing a good membrane model should be essential for further understanding of blood flow phenomena.

References

Effect of particle shape and size on vascular margination and wall adhesion: Design Implications in Drug Delivery

Michaela Cooley1, Apoorva Sarode2, Dmitry A. Fedosov3, Samir Mitragotri2, Anirban Sen Gupta1

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Micro- and nano-particle systems have emerged as important technology platforms for therapeutic delivery in the vascular and tissue compartments [1]. Functional efficacy of the particulate drug delivery systems is substantially influenced by their ability to marginate from the bulk of blood flow towards the vascular wall, in order to act at or beyond the wall. While the majority of particulate drug delivery systems are developed in the nanoscale size and spherical shape, several computational studies have indicated that nanoscale particles may face challenges regarding efficient margination in blood flow [2,3]. Computational studies have also indicated that in comparison to spherical particles, non-spherical shapes may have enhanced adhesion capabilities under hemodynamic flow environment [4,5]. To test these trends, several experimental studies have also been performed but often within a very limited range of relevant parameters. Rationalizing from such reports, we sought to substantially expand the evaluation metric by investigating the effect of particle morphology across a large size range (from 100 nm to 4.5 µm) and a large shape range (spheres, ellipsoids, rods, and disks) on their margination and wall-adhesion capabilities under simulated hemodynamic flow conditions at three hematocrit values (0, 20%, and 40% v/v). Experimental investigation was carried out using fluorescently-labeled polystyrene particles of various shapes and sizes, surface-coated with biotin and flowed over avidin-coated glass slide surfaces in a parallel plate microfluidic set-up at different shear rates and hematocrits. Experiments enabled the quantification of the number of particles adhered and retained per unit surface area of the chamber wall, under the various experimental conditions over time. In parallel, computational studies were performed to estimate particle margination and potential adhesion/retention capabilities for various conditions. Our results demonstrate that: (i) the presence of RBCs strongly influences particle adhesion at the wall across all sizes and shapes and (ii) a non-zero hematocrit significantly enhances the adhesion and retention of micro-scale particles of non-spherical shapes at the wall under flow. To this end, microscale particles of discoid shape seem to have the strongest performance. Future studies will explore additional effects of particle stiffness on their margination, surface-interactions and wall adhesion. Identification and validation of these parameters can lead to the design of superior performing particulate drug delivery systems compared to the traditional spherical nanoparticles.

References

Order and disorder in red blood cell traffic

Thomas Podgorski, Gwennou Coupier, Vassanti Audemar, Mehdi Maleki, Nicolas Rosuel, Zaiyi Shen, Chaouqi Misbah

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In the microcirculation, blood flows through a complex network of arterioles, capillaries and venules whose diameters typically range between 1-10 times the size of cells. The capillary bed is where most exchanges between blood and tissues occur as well as the place where the endothelial function regulates many phenomena under the influence of mechanical stresses and transported chemical fields, and therefore the details of the local blood flow and rheology.

From a structural perspective, the key features of microcirculatory blood flow are heterogeneity and self-organization controlled by hydrodynamic interactions coupled to cell mechanics and aggregation or adhesion forces.

At the scale of RBC traffic, this results in a non trivial distribution of the hematocrit, governs local apparent viscosity and stress fluctuations and leads to possible clogging/jamming phenomena in pathological cases [1]. Hematological and mechanical parameters govern elementary features such as structure-rheology coupling [1], separation at bifurcations [2] and dispersion phenomena [3,4], which triggers interest in possible biomedical applications.

References

Quantitative phase imaging (QPI) has emerged as an invaluable tool for imaging small transparent objects, such as biological cells and tissues. QPI employs various interferometric microscopy techniques to quantitatively measure the optical phase delay of samples. In particular, the measured optical phase delay provides information about the morphological and biochemical properties of biological samples at the single-cell level. Recently, QPI techniques have been widely applied to study the pathophysiology of various biological cells and tissues, including red blood cells (RBCs), white blood cells, bacteria, neurons, and cancer cells.

In this talk, we will present the recently developed 3-D holotomography setup using a dynamic mirror device, which is an optical analogous to X-ray computed tomography. In particular, we will discuss the visualization of 3D refractive index distributions of biological cells and tissues measured with the 3-D holotomography using the transfer function method, which has been widely used in the visualization field [1-4]. The outcome demonstrates outstanding visualization of 3D refractive index maps of live cells, which will be potentially used in various applications in biology and medicine. We will also discuss about the commercialization of the technique. In addition, we will also present the optical manipulation of eukaryotic cells on demands exploiting 3-D refractive index tomography.

References
Motion of a spherical capsule flowing in a branched tube with finite inertia

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A capsule is a liquid droplet enclosed by a thin membrane which can resist shear deformation. Capsules are widely found in nature in the forms of red blood cells (RBCs), eggs, …, but artificial capsules also have a vast range of applications in food, cosmetic, biomedical and pharmaceutical industries. In many situations, capsules are suspended in a fluid and flow through a complicated network of tubes or channels. Central to these flows is the path selection of capsules at bifurcations.

We computationally study the transient motion of an initially spherical capsule flowing through a right-angled tube bifurcation and entering through one of the entrances of the T-branch. It is modelled as a liquid droplet enclosed by a hyperelastic membrane following the Skalak law. Its motion and deformation are simulated using a three-dimensional immersed-boundary lattice Boltzmann method [1-2]. The fluids inside and outside the capsule are assumed to have identical viscosity and density. We mainly focus on path selection of the capsule at the bifurcation as a function of the parameters of the problem: the flow split ratio \( q \), the background flow Reynolds number \( Re \), the capsule-to-tube size ratio \( a/R \) and the capillary number \( Ca \), which compares the viscous fluid force acting on the capsule to the membrane elastic force. At low \( Re \), the capsule favours the branch which receives most flow. At higher \( Re \), inertia significantly affects the background flow: at equal flow split, a capsule tends to flow straight into the main branch and can still flow into it, even when the flow is much less flow than in the side branch. Increasing \( Ca \) promotes cross-stream migration of the capsule towards the side branch. We summarize the results in a phase diagram, showing the critical flow split ratio for which the capsule flows into the side branch increases with \( Re \). It shows that it depends on the size ratio and \( Ca/Re \) when the inertial forces are no longer negligible. This could be due to the bending of the fluid separation line towards the side branch.

The present results suggest that the trajectory of a capsule in a branched tube can be controlled by adjusting a range of parameters such as the capsule size, membrane elasticity, tube flow rate. One potential application of the results is to guide the development of microfluidic devices and use bifurcation geometries to enrich capsule suspension or sort deformable microparticles with different size or membrane elasticity.

References

The dynamics of blood flow in artificial microvascular networks

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Microfluidics technology provides a highly versatile platform for studying the dynamics of blood flow, and traffic of circulating cells and particles in complex networks of microfabricated vessels with the architecture inspired by the real microvasculature. When using these artificial microvascular networks, we can fabricate the desired geometrical configuration of the vessels in the network, ensure high-resolution imaging of cellular dynamics, and precisely control the properties and composition of the blood sample, while performing blood flow experiments at their natural scale, and under realistic flow conditions.

We have used this approach to show that (i) mechanical interactions with red blood cells (RBCs) could enable leukocyte entry into the blind-ended capillary sprouts, despite the unfavorable hemodynamic conditions and in complete absence of any chemoattractants [1]; (ii) spontaneous, self-sustained oscillations of capillary blood flow frequently observed in vivo can be generated solely by the non-linear rheological properties of blood flowing through microvascular networks, without any regulatory input [2]; and (iii) changes in RBC shape could significantly affect the ability of RBCs to perfuse microvascular networks, independently of other rheological parameters [3].

References

Stability of flowing red blood cell trains

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Red blood cells in sufficiently small vessels flow in approximately steady trains along the vessel center-line. However, in larger vessels the flow is more complex and apparently chaotic. This empirical behavior has been reproduced in detailed numerical simulations; here we analyze it as a linear stability problem. Extending a recent formulation for a two-dimensional model of this system \cite{1}, we analyze the asymptotic and transient linear stability of such red blood cell trains, including full coupling between the viscous fluid flow and cell membranes, which are represented as finite-deformation elastic shells. The linear system is constructed directly via imposition of an orthogonal set of small disturbances, with the corresponding hydrodynamic interactions evaluated with boundary integrals \cite{2}. Some disturbances are found to grow transiently (though decay for $t \to \infty$). These have relatively small-scale shape distortions and carry significant strain energy. In contrast, the $t \to \infty$ asymptotically unstable modes correspond predominantly to rigid-body-like displacements and rotations, and thus carry little strain energy. A range of intermediate cell–cell spacing is shown to be less unstable, particularly when the vessel diameter is relatively small. This is visualized in figure 1, along with examples of the associated disturbances. Increasing the viscosity ratio between cytosol and plasma from unity is destabilizing. Inflated and deflated cells are more or less unstable than a healthy red blood cell shape, depending on the train cell–cell spacing. Direct numerical simulations verify the stability predictions for small amplitudes and allow us to track the cell train into a disordered nonlinear regime.

\begin{itemize}
\item \cite{1} S. H. Bryngelson and J. B. Freund, Phys. Rev. Fluids, \textbf{1}, 033201, (2016)
\item \cite{2} C. Pozrikidis, (Cambridge University Press, Cambridge) (1992)
\end{itemize}

\begin{figure}
\centering
\includegraphics[width=\textwidth]{fig1.png}
\caption{(a) Asymptotic ($t \to \infty$) amplification $\alpha$ for a range of tube diameters $D$ and packing fractions $\phi$. (b) $\nabla_\alpha$ for cases (i)–(iv), magnified for visualization as $\vec{a} + a\hat{\varepsilon}\nabla_\alpha$ with $a\hat{\varepsilon} = 10$, which makes them visible at this scale though artificially ‘choppy’.}
\end{figure}
RATIONAL DESIGN OF NANOCONSTRUCTS FOR THE VASCULAR ADMINISTRATION OF THERAPEUTIC AND IMAGING AGENTS

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Over the last decade, multiple nanosystems have been developed for the diagnosis, imaging, and therapy of diseases, such as cancer, cardiovascular, chronic inflammatory and neurodegenerative. These nanosystems include nanoconstructs; microfluidic chips; nanosensors and actuators for medical devices. Most of these nanosystems have been developed following empirical approaches, while the notion of “rationally design” has been only recently realized. The use of computational modelling would certainly help in optimizing the biomedical performance of nanoconstructs for drug delivery and imaging. Indeed, as Computational Mechanics has already had a profound impact on science and technology, Theoretical and Computational Nanomedicine could have an equally pervasive impact in optimizing nanodevices and nanoconstructs for biomedical applications.

In this contribution, the author will present computational tools for the rational design of nanoconstructs meant to deliver with high specificity therapeutic and imaging agents upon systemic administration. in silico approaches for modelling the vascular transport, adhesion and extravascular diffusion of nanoconstructs and molecules will be reviewed, including the Isogeometric Analysis for describing vascular transport and adhesion of nanoconstructs in complex blood vessel networks; Immersed Finite Element Method and Lattice Boltzmann Method for analyzing the vascular and extravascular dynamics of nanoconstructs in microcapillaries. Challenges and opportunities for computational scientists in developing novel techniques for solving relevant problems in nanomedicine will be addressed.

ESSENTIAL REFERENCES
Vesicle dynamics and electro-hydrodynamics: modeling and computation

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In this talk, we introduce some numerical issues arising from 3D simulations for the vesicle problems. In particular, the major numerical challenges mainly fall into the geometrical modeling of 3D surface representations, and its computations on geometrical quantities and the derivatives along the surface. Instead of keeping the vesicle locally incompressible, we adopt a modified elastic tension energy to make the vesicle surface patch nearly incompressible so that solving the unknown tension (Lagrange multiplier for the incompressible constraint) can be avoided. Nevertheless, the new elastic force derived from the modified tension energy has exactly the same mathematical form as the original one except the different definitions of tension. We also introduce a hybrid immersed boundary (IB) and immersed interface method (IIM) to simulate the dynamics of a vesicle under electric field in Navier-Stokes flows. Within the leaky dielectric framework with piecewise constant electric properties in each fluid, the electric stress can be treated as an interfacial force so that both the membrane electric and mechanical forces can be formulated in a unified immersed boundary method. The electric potential and transmembrane potential are solved simultaneously via an efficient immersed interface method which incorporates the jump conditions naturally along the normal direction. The incompressible Navier-Stokes equations for the fluids are solved using a projection method on a staggered MAC grid and the potential is solved at the cell center. A series of numerical tests for the present scheme have been conducted to illustrate the accuracy and applicability of the method.

References


Many mechanical properties of the membrane of the human red blood cell (RBC) are already known. Under debate is the reference configuration (RC) of the shear elasticity. The return of the rim to its “home” location on the membrane after a transient mechanical manipulation of RBCs demonstrated that the RC is not spherically symmetric [1]. In most experiments sensitive to this lack of symmetry, the relevant indicators depend in addition on the shear stiffness and the 2D viscosity of the membrane. As a consequence several such experiments are required to extract the RC by fitting to appropriate computer models.

Observations on RBCs are usually performed in suspending media much more viscous than blood plasma. Subjecting such an RBC-suspension to shear flow, two basic modes are observed.

(i) Below a threshold in mean shear rate ($\dot{\gamma}$) and/or suspending phase viscosity ($\eta_0$) the RBCs rotate with little change in their resting shape. This state is referred to as tumbling (TB).

(ii) Above the threshold the RBCs assume a basically stationary orientation in the shear field while the membrane and the cell interior are in continuous rotation. This state is referred to as tank-treading (TT).

The following experiments are sensitive to the deviation of the RC from spherical symmetry.

threshold: Its dependence on $\dot{\gamma}$ and $\eta_0$ can be fitted to computer models.

**drift into C=0:** TB RBCs change orientation in such a way that their axis of rotational symmetry is parallel to the vorticity of the undisturbed shear flow [2]. The direction and the velocity of the drift can be compared to computer models.

**drift into C=$\infty$:** Irrespective of their initial orientation, TT RBCs change orientation such that after stop of flow and relaxation to the resting shape their symmetry axis is at right angles to the undisturbed vorticity. Again direction and velocity of the drift can be used.

**go&stop:** If TT is stopped with the rim displaced from its “home” position, RBCs will rotate in relocating the rim [1]. The velocity of this relocation can be used.

**SW and BR:** Superimposed on the stationary orientation during TT, RBCs display an oscillation of the angle of inclination and of the length of the long axis [3]. The amplitudes of both oscillations can be used.

**resting shape:** Provided the RC has rotational symmetry, a biconcave shape with a given ratio of dimple thickness to rim thickness can be generated in silico by choosing the spontaneous curvature of the membrane appropriately. Determination of the last would narrow the choice of possible RCs.

In modelling, traditionally stress-free-shapes (SFSs) have been assumed as RCs. Two cases were mainly considered: (i) the biconcave resting shape and (ii) oblate spheroids of variable eccentricity. However, evidence is accumulating that neither choice is compatible with all experimental data. Assuming that shear stresses relax in the long run, it is suggested to leave the concept of SFS in favour of a more general one in which the RC is not stress-free in any shape.

**References**


Contributed talk abstracts
Flow of healthy and thermally rigidified erythrocytes in a microfluidic rheometer

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For the first time, we report that the relaxation time \(\tau\) of healthy human Red Blood Cells (RBCs) decreases upon increase of both the buffer viscosity and its speed, and can reach values as low as 4 ms. Such result is almost two decades smaller than measurements usually reported in the literature using micropipette aspiration [1], ektacytometry [2] or microfluidics [3]. These published results (from 100 to 300 ms) are in good agreement with Hochmuth-Evans [1] analytical calculation of \(\tau\) using the Kelvin-Voigt model to describe the RBC as a viscoelastic material. They predict that \(\tau\) only depends on intrinsic cell membrane mechanical properties such as its viscosity and shear modulus.

We performed video-microscopic observations of healthy RBCs flowing in a microfluidic rheometer, consisting in a 5 µm high and 15 µm wide channel with oscillating width (varying 14 times from 5 to 25 µm and then back to 15 µm). A large range of fluidic stresses was investigated by varying both the buffer viscosity (\(\eta_{out} = 1\text{-}30 \text{ mPa.s}\)) by adding Dextran and the cell velocity (\(u_{RBC} = 100\text{-}2000 \mu\text{m/s}\)) through the modification of the applied pressure.

First we present two different modes of shape relaxation at the exit of the last constriction, according to \(\eta_{out}\) and \(u_{RBC}\). At high stress, as RBCs exit the last narrowing, they undergo a large stretching normal to the flow direction before relaxing to their equilibrium parachute-like shape; it is the stretching mode. On the contrary, in the unfolding mode - at lower stress, they relax directly from their passage in the last restriction and recover their equilibrium shape. We propose a theoretical analysis to elucidate the transition between the two relaxation behaviors, from a fluidic perspective, allowing the estimation of the shear elastic modulus \(\mu\) of RBCs in the range 4\text{-}10 \mu\text{N/m}, which is in good agreement with values reported previously in literature [4].

Then, we report how \(\tau\) decreases with increasing \(u_{RBC}\) and \(\eta_{out}\) [5], reaching values as low as 4 ms. Upon very low \(\eta_{out}\) and \(u_{RBC}\), we retrieve \(\tau\) similar to those reported in literature (\(\tau \sim 100\text{-}200\text{ ms}\)). We attribute the drop of cells relaxation time upon both parameters to energy dissipation. Finally, we compare the behavior of healthy and mechanically impaired RBCs flowing in our microfluidic rheometer, hence demonstrating its potential as diagnostic tool.

References

Red Blood Cells in a bi-dimensional channel: flow structure and cell rigidity impact

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In the vascular microcirculation RBC distribution is uneven in the direction normal to the blood flow, as first evidenced by the existence of a cell-free layer near the vessel wall [1]. In addition, the most rigid cells such as white blood cells and platelets are known to segregate to the walls while flowing in wide channels (a phenomenon named margination). A similar behavior is observed with rigid malaria-infected RBCs. While flow structure has been extensively studied theoretically, few experiments have been reported in which one-dimensional channels were used [2,3].

Here we develop microfluidic bi-dimensional channels (60 μm wide, 10 μm high, 5 mm long) in which RBCs are submitted to controlled flow and hydrodynamic stress. We study their collective behavior as a function of a number of physical parameters relevant for in vivo blood microcirculation such as RBC concentration, applied pressure drop through the device, channel width, and RBC rigidity. The flow structure is characterized using ultrafast video-microscopy and particle tracking.

We observe healthy RBCs, RBCs rigidified with glutaraldehyde, and mixture of healthy and rigidified RBCs. Initially dispersed healthy RBCs organize, while flowing along the channel, into aligned clusters. The cluster length depends on RBC concentration. On the other hand, rigidified RBCs do not cluster and mainly display tumbling motion as predicted for rigid disks [4]. Additionally similar measurements are performed on sickle RBCs in which the rigidity is increased due to a genetic mutation.

\textbf{Figure.} Healthy (A) and rigidified (B) RBCs flowing in 2D-channels.

References

Why do red blood cells roll in shear flow?

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As physiological blood flows are shear-dominated, a basic configuration in the study of hemodynamics is the one of isolated red blood cells subjected to pure shear flow. There has been a considerable amount of work to identify and understand the dynamics of isolated red blood cells in pure shear flow [1]. However, in spite of decades of research, this simple configuration is not fully understood yet, as demonstrated by the very recent discovery of new types of dynamics and shapes under flow in physiological conditions of viscosity and at high shear rates [2].

One of the striking dynamics of red blood cells occurs at low shear rates: red blood cells have been shown to flip in shear flow with an orbit that depends on the shear rate. Increasing the shear rate, the red blood cells exhibit an orbital drift, the orbits getting progressively closer to the rolling dynamics, where the small axis of the cell is aligned with the vorticity axis of the external flow. This orbital drift is yet to be fully understood, to be able to provide a complete picture of the dynamics of red blood cells in shear flow.

Using numerical simulations (http://www.math.univ-montp2.fr/ yales2bio/index.html), we show that in-plane membrane elasticity is the essential components to explain the orbital drift of red blood cells. The role of whole cell deformation can be separated and is shown to be a second-order effect. We also demonstrate that the frisbee motion [3] observed in vitro as a transient state may be stable under certain conditions and is again controlled by membrane in-plane elasticity. When the influence of membrane in-plane elasticity is decreased, we show that red blood cells in high-viscosity media roll while red blood cells in low-viscosity media are attracted by the shear plane and tank-tread and swing [1] as classically reported, which explains the dynamics of the cell when the shear rate increases. These findings are corroborated by a low-order model of the red blood cells dynamics.

References


Sorting red blood cells in deterministic lateral displacement devices

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Deterministic lateral displacement (DLD) method [1] for particle and cell sorting has attracted considerable research and technological interest for developing novel microfluidic devices for biological and clinical applications. As a size-dependent (label-free) particle separation method, the underlying mechanism is that the structure and geometry of DLDs uniquely determine the flow field, which in turn defines a critical particle size and the particle lateral displacement within a device. However, most bioparticles and cells of interest are non-spherical and deformable, and their shape and deformability are well known to largely influence their trajectories in DLD devices. Therefore, a rational design of such devices for an effective sorting of bioparticles requires a comprehensive understanding of the interplay between the dynamics and the deformation of soft particles in complex geometrical DLD structures.

In this work, we have performed mesoscopic simulations to explore possible sorting schemes of red blood cells (RBCs) in DLD devices [2, 3]. The dynamics and deformation of RBCs and the subsequent sorting have been intensively investigated by tuning the post shape, device geometry, cell rigidity, flow rate and viscosity contrasts between the intra-cellular fluid and suspending medium. In comparison to rigid spherical particles, the deformable RBCs exhibit much richer dynamics within a DLD device, which not only brings additional complications but also provides new separation mechanisms (e.g., based on cell dynamics or deformability), as shown in Fig. 1. The simulation results are very promising and demonstrate a great potential for the development of novel DLD devices to be used in medical diagnostics, since the change of cell deformability, morphology and dynamics takes place in many diseases (e.g., in malaria).

Fig. 1: Experimental (top) and simulation (bottom) trajectories of RBCs in DLD devices: (a) tumbling RBCs in the zigzag mode. (b) tank-treading RBCs in the displacement mode.

References
Red blood cells are highly deformable objects, which is a key factor for effective oxygen delivery to even the smallest blood vessels in our vascular system. Most strikingly, the cells can assume a wide variety of different forms when flowing through very small vessels. These include not only croissant and slipper shapes (as seen in figure 1), but also dynamic states such as tumbling motions.

Here we consider individual red blood cells flowing through a microchannel by means of optical experiments and fully three-dimensional numerical simulations. We show that the choice of the actually assumed shape does not only depend on intrinsic cell properties, but also on the initial condition. This especially includes the starting position relative to the channel’s center. We analyze this bistability in-depth and give the corresponding phase diagram.

**Fig. 1:** Two typical shapes of red blood cells flowing through microcapillaries. (a) Croissant and (b) slipper.
Impact of red blood cell rigidity on the vascular wall adhesion of neutrophils: implication in the pathology of sickle cell disease

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The symptoms of many blood related diseases can be attributed to irregularities in cellular dynamics that arise due to abnormalities in blood cells, particularly red blood cells (RBCs). Contingent on the disease and disease severity, RBCs can be afflicted with severe membrane rigidity. Extensive experimental research exists on characterizing the biochemistry of rigid RBCs in many diseases. However, little experimental work has been conducted towards isolating and investigating the effect of RBC rigidity on cellular dynamics, specifically on the segregation behavior known as margination and its effect on the binding functionality of other types of blood cells, i.e. white blood cells (WBCs) while in flow. We utilize an in vitro model to examine how different degrees of RBC rigidity and volume fraction of rigid RBCs, i.e. rigid RBC concentration, impact cell and particle margination, consequently impacting WBC and particle adhesion in blood flow. Additionally, we utilized confocal microscopy to gain a greater understanding of the effect of rigid RBCs on RBC core and cell margination. Fresh human blood used in all assays was obtained via venipuncture. RBCs were isolated from whole blood. Separated RBCs were treated with distinct concentrations of tert-Butyl hydroperoxide (TBHP) and reconstituted with healthy RBCs and plasma+WBCs at 40% final hematocrit for utilization in parallel plate flow chamber assays. RBC deformability was analyzed using a laser-assisted optical rotational cell analyzer (LORRCA; Mechatronics, Hoorn, The Netherlands). WBC and particle adhesion in blood flow with rigid RBCs is compared, i.e. normalized, to WBC and/or particle adhesion of healthy controls. Rigid RBCs are found to reduce WBC adhesion by up to 80%, contingent on the degree of rigidity and concentration of treated RBCs. WBC adhesion was reduced, although not always significantly lower than healthy control, in every iteration of the model, i.e. distinct wall shear rate (WSR) and RBC rigidities. Higher WSRs with rigid RBCs rendered a greater reduction in WBC adhesion. To compare RBC core distributions of different conditions to the healthy control with no rigid RBCs present, we used interquartile range (IQR) analysis and normalized to control. RBC core distribution IQR results show that the RBC core is expanded by up to 30% in size when rigid RBCs are present. These results hint that rigidity alone can largely disrupt normal hemodynamics and functionality of other blood cells.
Non-adsorbing macromolecules induce adhesion of diabetic red blood cells to the endothelium

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Abnormal adhesion of red blood cells (RBC) to endothelial cells (EC) has been linked to the pathophysiology of several diseases associated with vascular disorders. Various biochemical changes on the outer membrane of RBC as well as plasma protein levels, have been identified as being likely to play a key role, but the detailed interplay between plasma factors and cellular factors often remains unclear. In this work, we identified an alternative pathway by demonstrating that non-adsorbing macromolecules have a marked impact on the adhesion of diabetic RBC to EC.

We suspended RBC from patients with Type II Diabetes Mellitus (T2DM), in solutions of dextran to mimic the impact of non-adsorbing macromolecules. Static and continuous flow adhesion assays were used to determine the adhesion behavior of T2DM RBC with EC and the results compared with those of normal controls.

We found that the presence of non-adsorbing molecules in T2DM promote an increase in T2DM RBC - EC adhesion. It is concluded that this adhesion-promoting effect originates from macromolecular depletion interaction and thereby presents an alternative mechanism by which plasma proteins could regulate cell-cell interactions. These findings should thus be of potential value not only for a detailed understanding of the pathophysiology of diabetes mellitus but also other diseases associated with vascular complications.
A protocol for in vivo measurements of erythrocyte aggregation using ultrasound spectroscopy

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Erythrocyte aggregation is a non-specific marker of acute and chronic inflammation. Different laboratory instruments are available to assess the extent of erythrocyte aggregation but they require blood sampling and measurements such as low-shear blood viscosity or laser-based scattering and transmission aggregation kinetics. The aim of this study was to validate ultrasound spectroscopy imaging as an alternative to assess in vivo and in situ within blood vessels the extent of aggregation. Confounding variables such as the blood flow shear rate are affecting the level of aggregation and are increasing variability of ultrasound measures of inflammation. In this proof-of-concept study, we specifically aimed determining the robustness of an ultrasound erythrocyte aggregation measurement protocol for this application. Two techniques to reduce the impact of the blood flow were first evaluated with porcine and equine bloods circulated in a Couette flow device. These methods consisted in either stopping the flow or controlling it to a low value while acquiring ultrasound data. Then, repeatability and sensitivity of the retained control flow protocol were evaluated in eleven human subjects. Ultrasound measures at a mean frequency of 35 MHz were performed over the brachial vein and a pressurized device was used to reduce the flow velocity that was monitored with a speckle tracking algorithm. We observed that stopping the flow compromised interpretation and repeatability of the test as the initial state of flow affected aggregation measures based on spectral parameters extracted from the ultrasound backscatter coefficient. Conversely, maintaining a low flow provided repeatable measures and could distinguish between normal and high extents of erythrocyte aggregation. Excellent agreement was observed between the proposed ultrasound in vivo measures and ex vivo assessments of aggregation based on laser aggregometry (R² = 82.7%, p-value < 0.0001). These results support the feasibility of assessing erythrocyte aggregation in vivo in humans by quantitative ultrasound means.
RBC dynamics in microfluidic capillary models of organ-specific microvasculatures

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Microcirculation is generally organ-specific1,2 (e.g., lungs, mesentery, brain, kidney, liver, retina), where the local vascular morphology diverges significantly from long, straight vessels or simple dichotomous pathways. Pulmonary capillary networks (PCNs) embody a prime example of such organ-specific microvasculatures3, where blood vessels form dense meshes and notably, characteristic capillary lengths and diameters are similar to the size of individual red blood cells (RBCs). Inspired by the seminal “sheet flow” model that captures the assembly of such capillaries into quasi-2D networks4, we devise for the first time biomimetic microfluidic in vitro platforms of PCN structures perfused with RBC suspensions for various hematocrit levels (Hct). By implementing RBC tracking velocimetry, our measurements5 reveal a wide range of heterogonous RBC pathways that coexist synchronously within the PCN; a phenomenon that persists across the broad range of pressure drops (ΔP) and characteristic capillary segment sizes (dh) investigated. Interestingly, in spite of the intrinsic complexity of the PCN structure and the heterogeneity in RBC dynamics observed at the microscale, the macroscale bulk flow rate (Q) versus pressure drop (ΔP) relationship retains its linearity, where the hydrodynamic resistance of the PCN is to a first order captured by the capillary size dh. In a next step, we revisit the question of relative viscosity (μrel) at true scale for RBC suspensions in confined capillary networks and the role of Hct. Our findings not only corroborate with past experiments in scaled-up models6 but underscore for the first time how μrel is sensitive to the influence of dh. Overall, our in vitro efforts constitute a first, yet significant, step in exploring systematically the transport dynamics of blood in organ-specific microvasculatures.

References
An *in vitro* study of highly confined blood flows: From single bifurcations to 2D-networks

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Since the very first observations of microvascular networks in small animals by Jean-Marie Poiseuille in the XVIIIth century, the blood microcirculation has been extensively studied. One of the most striking feature highlighted by the French physicist is the highly heterogeneous distribution of the red blood cells (RBCs) throughout microvessel networks. Despite the intimate link between local RBC concentration (also called hematocrit) and surrounding tissue oxygenation, the coupling between microvascular architecture and hemodynamics is still poorly understood.

*In vivo* experiments provide data on spatio-temporal distribution of RBC concentration and velocities within a given microvascular network, but are limited to dilute regimes or highly confined flows, where the RBCs are arranged in single files and are therefore individually discernible [1]. Also, in such conditions, shape and diameters of the vessels cross-section is not precisely known. Along with physiological feedbacks, these uncertainties might be sources of errors. *In vitro* experiments can overcome such issues, inherent to the living [2]. Yet, providing reliable quantitative data at the scale of the blood microcirculation is challenging since the vessel diameters range from ~3 to ~10 µm, which is of the same order as the RBCs', or even smaller, and studies are often limited by the microfabrication process.

Our study aims at providing *in vitro* quantitative data on the distribution of the RBCs in geometries of increasing complexity: from diverging bifurcations to 2D-channel networks, with squared cross-section (WxW, W=5, 10, 20 µm). First, we have developed a calibration method that allows us to measure the hematocrit *in situ*, i.e. directly in the channel of interest, for a broad range of concentrations. Alongside with the hematocrit profiles, we are able to measure the RBC velocity profiles and thus deduce the RBC flow rate. By making simple assumptions on the suspending fluid, we deduce also the total blood flow rate. As a result, we have performed a parametric study of the phase separation (PS) effect, *i.e.* the non-proportional distribution of RBCs between the two daughter branches of a simple divergent bifurcation, and compared our results with the only *in vivo* empirically derived PS law [3]. Finally, we have designed 2D honeycomb networks and compared our experimental network perfusion with numerical simulations. For these networks, we show that the correlation between hematocrit and blood flow rate depends on the confinement of RBCs.

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**References**


Biomimetic bifurcating networks for studying RBC partitioning in microvasculature

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The dynamics of blood flow in microfluidic devices has been a subject of extensive research not only for RBC-plasma separation purposes but also to understand its complex behaviour at the microcirculation level. In particular, understanding the phenomena of RBC partitioning along consecutive bifurcations of blood vessels is crucial to explain the uneven haematocrit levels occasionally found in child branches, causing the lack of oxygen delivery to specific organs or tissues. Even though a few numerical studies for RBC modelling have focused on this problem [1] considering simple bifurcating geometries, a quantitative validation against experimental results is still needed.

The purpose of the present study is to systematically investigate the effect of distinct parameters, such as flow resistance, cell rigidity, angle of bifurcation and haematocrit level, on the RBC distribution in a bifurcating network. Some of the conditions tested experimentally are compared with numerical simulations using an immersed boundary-lattice Boltzmann 3D model [2], which takes into account the important effect of RBC deformability. The standard microfluidic geometry used, of constant depth and rectangular cross-section, is symmetric and composed by three consecutive generations of channels whose dimensions are initially defined using a biomimetic design rule [3], so the flow approximates real conditions found at the arterioles level.

The agreement between experimental and numerical data is fundamental to validate the 3D model for future studies on RBC flow in microfluidic devices. The information gathered in this work has the potential to help elucidate the blood flow dynamics in scenarios of channel deformability or cell rigidity [4], regularly related with common disorders like atherosclerosis or diabetes, respectively.

References

Possible Self-margination of sickle red blood cells suspensions in glass capillaries

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When blood is flowing, red blood cells (RBCs) axial migration influences the radial distribution of the other circulating cells, such as white cells and platelets [1–4], a phenomenon known as margination. The ability for healthy RBCs to deform and to aggregate may influence such axial migration [5]. In sickle cell anemia, RBCs population is very heterogeneous in terms of density and rigidity/deformability compared to healthy cells. Then, a natural question arises: do sickle RBCs self-marginate? With this question in mind, the collective behavior of oxygenated sickle RBCs and their distribution along cylindrical micro-capillaries with diameters comparable to a human venule or arteriole was investigated. Our results show that there is indeed a heterogeneous distribution of cells according to their density: low-density cells tend to stay closer to the center of the channel, while most dense cells (also more rigid) self-marginate under defined conditions. This self-margination heterogeneity could influence the ability of some sickle RBCs to adhere to the vascular wall and slow down blood flow. Aggregation seems to inhibit self-margination depending on the aggregative factor and patient: dextran allows self-margination in some patients and inhibits it in others. Plasma inhibits self-margination of cells in all cases, highlighting the importance of the plasma proteins and adhesive molecules in the aggregation phenomena.

References

Experimental ultrasound characterization of red blood cell aggregation: estimation of the aggregate size distribution

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Quantitative ultrasound techniques for determining red blood cell (RBC) aggregate structures rely on a theoretical scattering model to fit the BackScatter Coefficient (BSC) of blood to an estimated theoretical BSC. The scattering theories commonly used are the Structure Factor Size Estimator (SFSE) and the Effective Medium Theory (EMT) combined with the Structure Factor Model (EMTSFM), that assume locally RBC aggregates of identical size [1] [2]. The aim of this work was to further develop the EMTSFM to incorporate the polydispersity in terms of aggregate size, and assess its ability to estimate the aggregate size distribution with in vitro experiments.

We propose two successive experiments: a first experiment on blood sheared in a Couette flow device coupled with an ultrasonic probe, and a second experiment, on the same blood sample, sheared in a plate rheometer coupled to a light microscope. The ultrasound study showed that the polydisperse EMTSFM is the most appropriate model for characterizing red blood cell aggregates. The polydisperse models provide the best fit to the experimental BSC data (when compared to the classical monodisperse models). Lastly, satisfactory correlation is obtained ($r^2 \approx 0.92$) between the mean aggregate size estimated with ultrasound and optics.

![Image of experimental setup and data analysis](attachment:image.png)

**Fig. 1:** (a) Couette flow device combined with an ultrasound probe. (b) Frequency dependent backscattering coefficient, measured (symbols) and fitted with EMTSFM polydisperse theory (solid lines). (c) Comparison between probability density functions obtained by ultrasound and optics.

**References**

Microvascular networks in the human body are made of the smallest blood vessels, namely, the capillaries, arterioles, and venules. High-fidelity in silico modeling of blood flow in microvascular networks, however, remains a major challenge. Blood in small vessels behaves as a concentrated suspension primarily comprised of deformable red blood cells (RBC). The architecture of vascular networks is complex, and characterized by bifurcating, merging, and winding vessels. Furthermore, the network topology varies from organ to organ. Such geometrical differences may result in significant deviations in hemodynamics in a long straight tube versus a vascular network.

We have developed a direct numerical simulation method for cellular-scale blood flow in physiologically realistic microvascular networks. The model resolves large deformation of deformable blood cells flowing in arbitrarily complex geometry. The vascular networks are constructed in silico following published in vivo images and data, and are comprised of bifurcating, merging, and winding vessels. The vascular walls are modeled using a sharp-interface ghost node immersed boundary method (IBM), while the deformable interfaces of the RBCs are modeled using a continuous forcing IBM. Each of these components is seamlessly integrated into the framework of a coupled finite-volume/spectral flow solver. A continuum description of the RBC is used by representing it as a viscous drop made of hemoglobin surrounded by a zero-thickness membrane, and a finite-element method is used to compute the membrane tension.

Several unexpected results are observed in our simulations: in several vessels we find that the flow rate and pressure drop are negatively correlated; the flow resistance and hematocrit are also negatively correlated. We observe cell jamming at vascular bifurcations leading to large temporal ‘spikes’ in flow resistance [2]. Disproportionate partitioning of cells with respect to flow partitioning at vascular bifurcations is also observed. Interestingly, however, we find that cell partitioning at some bifurcations is disproportionately less than the flow partitioning, suggesting a reverse Zweifach-Fung effect, due to the influence of sequential bifurcations.

References

Modelisation of blood perfusion into a whole adipose tissue vascular network

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Fat subcutaneous tissue is a densely vascularized parenchyma, the micro-vascular structure of which has been poorly documented. It is one of the few adult mamal tissue known to exhibit great plasticity with the ability to quickly adapt to various physiological conditions. It has been shown than a 'core region' close to the lymphatic ganglion is the site of an heterogeneity in the tissue structure as it is composed of dense aggregates of cells and vessels called 'lobules'[1]. Lobular structures exist in the liver and their homeostatic role of which has been demonstrated [2]. Surprisingly very few studies have investigated similar functions of adipose tissue lobules.

In order to analyse the micro-vascular structure of adipose tissue, we developed a new tissue preparation and image acquisition protocol using Light Sheet Microscopy (LSM). Using adapted vessel segmentation image analysis methods, we are able to reliably extract and reconstruct the entire vascular network, comprising up to 1.6 millions of vascular segments, as illustrated in Figure 1. Since segmentation permits a robust estimation of hydraulic structural parameters such as local vessel’s diameter and tortuosity, it allows blood flow perfusion evaluation using resistance network models [4, 3]. One advantage of such an exhaustive micro-vascular reconstruction over an entire tissue, is to avoid cutting vessels at the edge of the reconstructed volume, avoiding unspecified boundary conditions. In this work, we quantify the hydrodynamic couplings between lobules, and show how heterogeneous perfusion displays within fat tissue.

Fig. 1: Results of the vessel’s segmentation procedure on a full 3D inguinal adipose tissue of a mouse imaged by LSM. The original tissue size is 3 cm³. a. displays the architecture of large vessels only, b. shows the entire micro-vascular network comprising (1.6 million vessels), c. and d. illustrates focus on specific regions.

References
Deformability-influenced delivery of red blood cells in microvascular networks

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In vivo, blood flows through a complex network of the microcirculatory system in order to ensure oxygen delivery and remove metabolic waste. Understanding blood flow in microvascular networks and its dependence on the mechanical properties of its constituents are essential to understand some physiological implication in microcirculation. In real situations, the vascular network consists of many short vessel segments with length of about hundreds micrometers¹ which are not long enough to sufficiently allow development of the blood flow, especially in the dilute case. The corpuscular nature of the red blood cell (RBC) distribution together with the fact that the flow is not always fully developed in short enough vessels make the problem quite challenging and large deviations can be manifested as compared to the classically adopted picture.

To better understand the blood flow properties in the microcirculation, direct numerical simulations of RBC suspensions in a 2D network are carried out². As a first step we consider the network to be ordered, a choice dictated by reducing complexity in order to clarify the sole effect of the branching. A structured network with hexagonal loops is used to make a symmetrical flow at each bifurcation. We examine the effect of the multistage bifurcation. The motion of single RBC in the network is firstly investigated. We shall see that the configurations of RBC in the downstream position depend on the previous states of RBC in the upstream position. This results in a quite rich behavior when the RBC meets a bifurcation. For example, a chaotic partition of RBC may take place at bifurcations, and this clearly has an impact on the overall displacement of RBCs in the network. We find that the deformability of RBCs impacts the displacement for low concentrations, up to about 20%, beyond which the crowding effects and cell-cell interactions make no big distinctions among different RBC deformabilities. Besides the above effects, we discover that RBCs exhibit larger flux in the network when they are more rigid and this is in a marked contrast with the scenario in straight tubes. Finally, we will see that cells enjoy a faster longitudinal diffusion when they have a smaller deformability.

References
Red blood cell passage of small capillaries is associated with transient Ca\textsuperscript{2+}-mediated adaptations

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When red blood cells (RBCs) pass constrictions or small capillaries they need to pass apertures falling well below their own cross section size.

We used different means of mechanical stimulations (hypoosmotic swelling, local mechanical stimulation, passing microfluidic constrictions) to observe cellular responses of human RBCs in terms of intracellular Ca\textsuperscript{2+}-signalling by confocal microscopy of Fluo-4 loaded RBCs. We were able to confirm our \textit{in vitro} results in a mouse dorsal skinfold chamber model showing a transiently increased intracellular Ca\textsuperscript{2+} when RBC were passing small capillaries \textit{in vivo}. Furthermore we performed the above-mentioned \textit{in vitro} experiments as well as measurements of RBCs filterability under various pharmacological manipulations to explore the molecular mechanism of the Ca\textsuperscript{2+}-signalling.

Based on these results we conclude that mechanical stimulation of RBCs activates mechanosensitive channels like Piezo1. This channel activity allows Ca\textsuperscript{2+} to enter the cell, leading to a transient activation of the Gardos channel associated with K\textsuperscript{+}, Cl\textsuperscript{-} and water loss, i.e. with a transient volume adaptation facilitating the passage of the RBCs through the constriction.
Production of blood platelets in a microfluidic chip

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Platelets are small anucleate cells that circulate in blood and are responsible for the arrest of bleeding. Platelets are formed by fragmentation of larger cells called megakaryocytes. Thrombocytopenia (insufficient platelet count) is a major condition, often requiring platelet transfusions. High collection costs, donor shortage, immunogenicity, risk of contamination and storage issues represent the main limits of this therapeutic approach.

We present a new, rapid method for producing blood platelets in vitro from cultured megakaryocytes based on a microfluidic device. This device consists in a wide array of von Willebrand factor-coated micropillars. Such pillars act as anchors on megakaryocytes, allowing them to remain trapped in the device and subjected to hydrodynamic shear. The combined effect of anchoring and shear induces the elongation of megakaryocytes and finally their rupture into platelets and proplatelets in the flux. This original bioreactor design allows to process megakaryocytes at high throughput (millions per hour). Since platelets are produced in such a large amount, their extensive biological characterization is possible and shows that platelets produced in this bioreactor are functional [1].

Reference

Tissue growth pressure drives early blood flow in the chicken yolk sac

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Understanding how molecular and physical cues orchestrate vascular morphogenesis is a challenge for developmental biology. Only little attention has been paid to the impact of mechanical stress caused by tissue growth on early blood distribution. Here we study the peripheral accumulation of blood in the chicken embryonic yolk sac, which precedes sinus vein formation.

We report that blood accumulation starts prior to heart-induced blood circulation. We hypothesized that the driving force for the primitive blood flow is a growth-induced gradient of tissue pressure in the yolk sac mesoderm. Therefore, we studied embryos in which heart development was arrested after two days of incubation, and found that yolk sac growth and blood peripheral accumulation still occurred. This suggests that tissue growth is sufficient to initiate the flow and the formation of the sinus vein, whereas heart contractions are not required. We designed a simple mathematical model which makes explicit the growth-induced pressure gradient and the subsequent blood accumulation, and show that growth can indeed account for the observed blood accumulation.

This study shows that tissue growth pressure can drive early blood flow, and suggests that the mechanical environment, beyond hemodynamics, can contribute to vascular morphogenesis.
Mesoscale modeling of blood clot contraction

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Blood vessel injury triggers a series of physiological responses that are culminated by the formation of a clot arresting the bleeding. Blood clotting disorders may prevent the body’s natural ability to achieve hemostasis. This can result to a variety of life threatening conditions such as, excessive bleeding, stroke, or heart attack. Understanding of the underlying physics behind the clotting process is critical for effective treatment of these and other blood disorders. Clotting is a highly complex multi-scale process involving platelet activation, polymerization of fibrin networks, and development of platelet-fibrin hemostatic plug that retracts to facilitate normal blood flow. We develop a mesoscale model based on dissipative particle dynamics to get fundamental insights into the dynamic process of clot contraction. We use image analysis to extract characteristics of fibrin network and platelet distribution from 3D confocal microscopy images of clot time evolution. Using experimental estimates of the crosslink density in the fibrin networks and platelet locations, we create computational model of fibrin-platelet network and use this model to probe platelet-fibrin interactions during clot retraction. We examine how the clot evolves in time and compare the results with experimental observations to validate our mode. We probe clot contraction when it is constrained by two opposing walls and examine the morphological changes in the fibrin network and the platelet distribution and clustering. We also use our model to examine the effect of different platelet parameters on the contraction process and clot micromechanics.
Platelet activation in intensive blood flows

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Mechanisms of platelet sensitivity to blood shear stress is discussed, in the framework of far-from equilibrium approach to the analysis of vWF conformational unfolding on the platelet surface [1]. It is demonstrated that the degree of unfolding of vWF macromolecule depends on the number of it’s monomeric units – N as well as on the intensity of shear stress – τ.

Supposing that platelet is activated only in the case when some overcritical number of monomers – (N > N_A) are simultaneously connected with surface platelet receptor GPIbIIa, explicit formula for platelet activation risk index is derived. Relevant phase diagram as well as bifurcation diagram are presented.

Platelet activation under intensive flow conditions may be treated as a kind of far-from equilibrium phase transition of the second order.

References

A physical description of platelet deposition

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We have recently developed\cite{1} a numerical model based on first principle physical mechanisms to explain the adhesion and aggregation of platelets subject to a shear induce diffusion. The model was developed in synergy with in vitro experiments performed with the so-called Impact\textsuperscript{R} device\cite{2}. The experiments use whole blood from healthy donors. It gives, the number of deposition aggregates, their average size, the amount of activated and non-activated platelets still in suspension at times 0\,s, 20\,s, 60\,s, 120\,s and 300\,s. Our model can reproduce very well the experimental findings, as illustrated in Figure 1.

However our study reveals several mechanisms that are poorly understood or neglected in current literature. In particular: (1) we could determine the thickness of the aggregates, and measure the volume of a deposited platelet\cite{3}; (2) we observed and quantified the role of albumin as an inhibitor of platelet deposition; (3) we clarified the role of pre-activated and activated platelet in the deposition process; (4) we showed that the usually accepted shear induced diffusion coefficient is much to small to explain the observed platelet deposition rate, thus suggesting different transport mechanisms; (5) by comparing the experiments with the numerical simulations we can infer the adhesion and aggregation rates and other relevant parameters.

\begin{figure}[h]
\includegraphics[width=0.4\textwidth]{cluster.png}
\caption{Clusters of platelets formed on the deposition surface of the Impact\textsuperscript{R} device. Left: experimental result. Right: numerical simulation with properly tuned parameters.}
\end{figure}

References


Cell-based platelet aggregation modelling in Hemocell

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Thrombosis is a complex problem that may lead to a stroke or a heart attack. Many continuous models are developed to study thrombosis [1]. However, the question arises if the continuous approach is valid or a more detailed cell-based approach is needed. Our goal is to build a cell-based platelet (PLT) plug model on top of Hemocell, a high performance microscopic cellular library. Cell-based thrombus models found in the literature lack of red blood cells (RBCs) and use incorrect PLT-shapes [4]. We start working on our platelet plug model by obtaining a better understanding of the role of the von Willebrand factor (vWF) during the adhesion of platelets to the wall and during the formation of a platelet aggregate.

To study the behavior of RBCs, PLTs and the vWF in an aggregation favoring area, an experiment of Nesbitt et al. (2009) [2] is used as test case (see figure 1a). In this experiment glass spheres of different diameters (2, 5, 9 and 15 µm) coated with vWF are fixed on the bottom of a flow channel that was perfused with whole blood. From this experiment, we can conclude that RBCs form a cell-free layer in the region where in the experiments the platelet aggregate is formed (see figure 1b). This area is dependent on the diameter of the sphere and the hematocrit of the blood. Additionally, the elongation of vWF is studied in the area behind the sphere. We hypothesize that vWF can only elongate properly in absence of RBCs and under certain shear conditions. Additionally, the concept of elongational flow in relation to vWF is worth to study [3]. The goal of this experiment is to predict the probability of platelets to enter the RBC-free region and their probability to bind to vWF.

Figure 1: Experiment with sphere diameter of 15µm: (a) Nesbitt’s [1] experiment (b) simulation with Hemocell with a RBC-free region length of 71.7 lattice units (35.9 µm).

References
Red blood cell rheology and vascular dysfunction in sickle cell disease

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Sickle cell anemia (SCA) is a genetic disease characterized by the presence of abnormal hemoglobin (HbS) that polymerizes under deoxygenated conditions causing a mechanical distortion of red blood cells (RBC). SCA patients have decreased RBC deformability and increased RBC aggregates strength. We recently investigated the contribution of blood rheology and vascular dysfunction in vaso-occlusive crises (VOC). Our findings demonstrated that SCA patients have blunted microvascular reactivity during local thermal heating tests compared to controls. The lower microvascular reactivity was negatively associated with the levels of plasma advanced oxidation protein products and nitrotyrosine suggesting a key role of oxidative/nitrosative stress in vascular dysfunction in this disease. Moreover, we recently observed that circulating exosomes in SCA, originating mainly from RBCs, were able to promote monocytes adhesion to endothelial cells through an increase in P-selectin expression and alter in vitro endothelial cells barrier permeability and the topographic distribution of the tight junction protein ZO-1 in a SCA severity-dependent manner compared to healthy children. These new data suggest that exosomes originating from RBCs could be one of the sub-cellular elements involved in the endothelial dysfunction associated with SCA. Finally, multivariate analyses recently performed in SCA cohorts showed that increased blood viscosity and decreased microcirculatory oxygenation are independently associated with a higher risk to develop frequent VOC episodes. In conclusion, vascular dysfunction and blood hyperviscosity emerge as key factors involved in the severity of SCA and the occurrence of frequent VOC events.
Physical investigation of hemo-rheological characteristics for cardiac surgery patients

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We employ high sensitivity physical measurements of blood viscosity and elasticity to examine differences between healthy people and patients who underwent cardiovascular surgery. Cardiovascular diseases (CVD) and heart-related complications mortality have remained the main cause of death worldwide at this time[1]. However, current indicators for rehabilitation are both expensive and time-consuming.

Coronary Artery Bypass Graft (CABG) and Mitral Valve Replacement (MVR) surgeries are widely accepted treatments for advanced atherosclerosis and coronary artery disease. There are high risks related to post-operative complications such as cerebral ischemia, systemic inflammation, deep vein thrombosis, and endothelial injury, etc. However, there are currently few indicators to monitor.

Hemorheology is an established tool for monitoring the blood perfusion efficacy and health care for surgical patients. We recruited twenty-seven CVD patients (N=27) who needed the cardiopulmonary bypass (CPB). Each assessment involved a baseline recording before surgery (M0), one day(M1), one week(M2) and one year(M3) following surgery. The viscosity and elastic modulus were measured by using a Physica Rheometer MCR 501(Anton-Paar, Graz, Austria)[2]. Moreover, the life-quality questionnaire also recorded the patients’ habits (smoking habit, diastolic blood pressure, and low-density lipoprotein cholesterol).

We found the increase in whole blood viscosity (WBV) correlated with elevated C-reactive protein and plasma fibrinogen levels[3]. Blood viscosity was found to be higher for the patients than for the healthy control group. WBV were also elevated in the post-surgical group(M1, M2) than the pre-surgery group (M0), possibly due to acute-phase response for surgery intervention. However, WBV returned to baseline in one year after surgery (M3)

References

Uncertainty quantification of the Red Blood Cell model

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Large Data, advanced sampling methods and High Performance Computing are actively changing the landscape of computational life sciences. We integrate these components under a Bayesian uncertainty quantification (UQ) and prediction framework [1] and apply it to study the mechanics of Red Blood Cells (RBCs).

Being the key constitutive component of blood, RBCs have always been subject of interest in various experimental and, lately, computational studies. However, employed computational models of erythrocytes are semi-empirical and contain a number of parameters to be identified. The common practice is that these parameters are calibrated with simplifying assumptions and using the averaged experimental data without its uncertainty. Therefore such parametrization is unable to provide a range, or a distribution of parameters that is necessary for studies of collective behavior of many cells.

Our show the Bayesian approach to the calibration of the RBC model based upon the Dissipative Particle Dynamics (DPD). Combining experimental studies from the literature and from the group of Prof. Mauro Ferrari, we quantify uncertainties of the membrane model using data on RBC stretching and its rotation frequency in the shear flow and compare the obtained parameters with those used in literature. Furthermore, we propagate the uncertainties in the blood rheology simulations involving thousands of cells.

We employ uDeviceX [2] (ACM Gordon Bell Prize Finalist 2015), a high-throughput software designed for predictive simulations of microconfined blood flows at large scales using state-of-the art DPD (Dissipative Particle Dynamics) models. Targeted and carefully optimized for GPU-enabled supercomputers, uDeviceX is capable of unprecedented simulations of blood flows at sub-micron resolution in realistic geometries.

References
Hydrodynamics of pulsatile flows

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Pulsatile flows occur in a variety of engineering applications and most notably in the human body. While many hydrodynamic studies have addressed the behaviour of purely oscillating flows (with no mean-flow component), surprisingly few investigations have considered the fundamental dynamics of pulsatile flows, even in simple geometries. Hence, the purpose of the present work is to contribute to the understanding of essential hemodynamic flow properties by systematically characterizing the dynamics of small-amplitude perturbations as well as the fully developed flow features prevailing in pulsatile channel flows [1].

The time-periodic base flows are known analytically and completely determined by the Reynolds number $Re$ (based on the mean flow rate), the Womersley number $Wo$ (a dimensionless expression of the frequency) and the flow-rate waveform.

Linear stability characteristics are obtained both by Floquet analyses and by linearized direct numerical simulations. In particular, the long-term growth or decay rates and the intracyclic modulation amplitudes are systematically computed. At large frequencies (mainly $Wo \geq 14$), increasing the amplitude of the oscillating component is found to have a stabilizing effect, while it is destabilizing at lower frequencies; strongest destabilization is found for $Wo \simeq 7$. Whether stable or unstable, perturbations may undergo large-amplitude intracyclic modulations; these intracyclic modulation amplitudes reach huge values at low pulsation frequencies.

For linearly unstable configurations, the resulting saturated fully developed finite-amplitude solutions are computed by direct numerical simulations of the complete Navier–Stokes equations. Essentially two types of nonlinear dynamics have been identified: ‘cruising’ regimes for which nonlinearities are sustained throughout the entire pulsation cycle and which may be interpreted as modulated Tollmien–Schlichting waves, and ‘ballistic’ regimes that are propelled into a nonlinear phase before subsiding again to small amplitudes within every pulsation cycle. Cruising regimes are found to prevail for weak base-flow pulsation amplitudes, while ballistic regimes are selected at larger pulsation amplitudes; at larger pulsation frequencies, however, the ballistic regime may be bypassed due to the stabilizing effect of the base-flow pulsating component.

By investigating extended regions of a multi-dimensional parameter space and considering both two-dimensional and three-dimensional perturbations, the linear and nonlinear dynamics are systematically explored and characterized. A particularly important part of this investigation is the detailed characterization of the hydrodynamic forces acting on the fluid-containing boundaries. These results pave the way for taking into account physiological pulse-forms and more complex geometries.

References

Rolling adhesion of malaria-infected red blood cells

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Malaria in humans is caused by the eukaryotic parasite \textit{Plasmodium falciparum}. The clinical symptoms occur mainly during the blood stage of infection which proceeds in three stages: (1) ring stage, (2) trophozoite stage and (3) schizont stage. During these stages, the parasite increasingly remodels the red blood cells, mainly by modifying the spectrin network and forming a system of adhesive protrusions called knobs. These knobs make the infected cell to stick to endothelial cells and causes capillary obstruction. First we will discuss how exactly the shape of infected red blood cells changes during the whole infectious cycle. For this purpose we have measured volume and surface area as a function of time post infection using confocal microscopy and image processing [1]. We will then discuss results concerning rolling adhesion of trophozoites and schizonts using flow chamber experiments [3]. With the help of adhesive dynamics simulations [3] and deformable cell model [4, 3], we show how the shape and stiffness of these cells contribute to different rolling behavior such as flipping or rolling adhesion. We will also discuss how details of knob structure such as density or multiplicity factor affect the adhesive dynamic phase diagram.

References


Intermediate regime and a phase diagram of red blood cell dynamics in a linear flow

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Dynamics of a single red blood cell (RBC) has been a subject of intensive research due to its relevance to blood rheology and pathologies related to a blood flow. Despite the recent experimental numerical and theoretical advances on RBC dynamics in shear flow the following problems remain unresolved: (i) existence of the intermediate regime and its dynamical signature, (ii) shape deformations in the well-studied swinging regime, (iii) the RBC stress-free shape, and (iv) phase diagram of RBC dynamical regimes. We address these questions in a unique experimental approach, namely the dynamical trap in a microfluidic 4-roll mill device that allows long time observations of a single RBC in a linear flow in all dynamical states (Figure 1). This allows us (i) to construct a phase diagram of RBC dynamics in a linear flow, (ii) to study in details the RBC dynamical states, and (iii) to characterize the intermediate, stable regime between the well-known tumbling and swinging with surprisingly large shape deformations (Figure 2). Finally, together with complementary experiments in trapping extensional flow and comparison with several numerical models we conclude that the biconcave stress-free shape is a more probable one.

Fig. 1
Fig. 2

References
Soft biolubrication: lift forces at a vascular wall mimic

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Interactions between circulating cells and blood vessel walls are central to many physiological processes, such as the early stages of the immune or inflammatory response, gas exchanges with tissues, or vascular remodeling. Among these interactions, hydrodynamic forces play a key role, as they control the radial migration of the circulating cells towards or away from the vascular walls. A well-known example of hydrodynamics at play is the so-called cell-free layer, a near-wall region that is depleted in red blood cells, which is classically interpreted as resulting from the non-inertial repulsive force arising between the vessel surface and the deformable red cells flowing nearby [1]. A point that has been largely overlooked so far is the contribution of the wall deformability to such a repulsive hydrodynamic force, and in particular the role of the endothelial surface layer, a compliant meshwork of glycosaminoglycans lining the lumen of blood vessels.

In this context, we have developed an experimental setup allowing for the study of microparticles flowing past a surface bearing a macromolecular layer of hyaluronic acid mimicking the nature, thickness (a few hundreds of nm) and elastic properties (~100 Pa) of the endothelial surface layer. Combining parallel plate flow assays and 3D particle tracking based on interference microscopy, we show that:

(i) non-deformable spherical microbeads traveling, in a controlled shear flow, close to the macromolecular layer, are repelled and lift away from the surface under strong enough (yet physiological) shear rates,

(ii) the bead/surface distance increases with increasing shear rate and can reach up to several hundreds of nm,

(iii) our experimental results can be quantitatively described in the theoretical framework of elastohydrodynamics accounting for the effect of substrate deformations [2].

This represents, to the best of our knowledge, the first experimental demonstration that a thin and compliant surface layer can contribute substantially to the repulsive hydrodynamic interactions with flowing, cell-sized objects.

References

3D tomography of red blood cells in micro-channels

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We propose a new confocal 3D imaging technique for fluorescent stained red blood cells (RBCs) in micro-fluidic flow. Our approach allows us to recover the full 3D representation of moving RBCs under conditions prevailing in the micro-vasculature. As key feature, we employ a micro-fluidic channel which is tilted by a small angle with respect to the objective. This forces cells to pass the focal plane in an inclined manner and a stack of cross-sectional images is recorded for each traversing object. Image slices are then assembled to recover the volumetric representation of individual cells. In contrast to common scanning approaches, the present method relinquishes any mechanical actuation of the objective or stage and frame rates up to 600 FPS can be realized since no mechanical delay is involved. At maximum frame rate, cells up to a velocity of 1.5 mm/s can be recovered. Even if our approach appears straightforward, a number of sophisticated image processing steps are necessary for successful 3D recovery. For instance, optical artifacts from the spinning disk at high frame rate must be compensated for a smooth reconstruction. Moreover, the cell velocity must be determined very precise to achieve a correct stacking of individual image slices. In a micro-channel of 25 µm × 10 µm, we were able to find two equilibrium cell shapes under certain flow condition: the ‘slipper’ and the ‘croissant’ shape (Fig. 1). Validating this result, numerical simulations are performed which are in good agreement with experimental observations.

Figure 1: 3D measurements (a) and simulation results (b) of RBCs in a micro-channel of 25 µm × 10 µm cross-section. Cell velocities correspond to 330 µm (slipper), respectively 370, µm (croissant).
Poster abstracts
Margination of blood cells

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Due to their unique shape and mechanical properties, red blood cells (RBCs) undergo a so called lift force in the flow pushing them away from the vessel wall. It leads to creation of "cell-free layer" (CFL) at the periphery of the flow. The CFL has lower viscosity compared to the core of the blood flow which serves as a lubrication, facilitating the passage of blood in microvasculature. And while RBCs have tendency to move towards centerline of the flow, other blood components, such as white blood cells (WBCs) and platelets, migrate towards walls of blood vessels. This process is commonly referred to as margination. Margination in blood is a phenomenon of great physiological importance and can have applications in cell separation and targeted drug delivery. However, precise mechanisms involved in this process remain unclear. Margination appears to be a complicated process depending on many parameters such as size of the marginating particles, their shape, volume fraction of RBCs and etc.

In our research we study rigidity based margination in binary suspension of deformable and stiff particles. For this purposes we observe and quantify blood flow consisting of 2 populations of red blood cells - healthy and rigidified with cross-linking agent (glutaraldehyde) - in microfluidic channels in case of different flow rates, hematocrits and vessel geometries. Using such experimental model allows us to examine margination caused exclusively by rigidity contrast between to subpopulations of particles. In contrast to previously performed experimental studies where imaging of marginating particles was conducted by adjusting the focal plane in the middle of the microchannel, we used confocal microscopy to reconstruct 3D distribution of labeled cells across the section. In our work we accent following topics: (i) We investigate margination of rigidified RBCs in microchannels of different geometries and we demonstrate possible margination paths in rectangular channels, cylindrical capillaries and confined microchannels, imitating pseudo 2D blood flow. (ii) We show how distribution of rigid cells changes in the channels lengthwise and how margination reaches the saturation for different hematocrit levels. (iii) Effect of varying flow rate was studied for wide range of values and we conclude non-linear relationship between velocity and margination level.
In vitro measurements of apparent intrinsic viscosity in function of tube hematocrit and red blood cell velocity

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In microvascular networks, local red blood cell (RBC) velocity and tube hematocrit (Hₜ) affect the apparent viscosity and pressure/flow fields in the whole network. To our best knowledge, studies of the apparent intrinsic viscosity (Kₜ) are mainly computational [1,2] and only few in vitro studies are available [3]. The aim of the present study is to conduct experiments in a single microchannel to quantify the apparent intrinsic viscosity of blood in function of the RBC velocity and hematocrit. The microdevice made from PDMS had a single straight channel of 500µm length with square cross-section (8µm x 8µm). RBC suspensions of porcine blood were prepared as described in [4] with two different base hematocrits (2.5% and 5%). To drive the flow, two different hydrostatic pressure heads between the fluid level in the reservoir and the outlet were set (4.5 cmH₂O and 7.5 cmH₂O). RBC velocities and tube hematocrits were determined from line scans stacked in temporal sequence [4]. These values were then used to calculate the apparent intrinsic viscosity. The mean value of Kₜ (1.79) is in the range of the data reported in numerical studies [1,2]. We found a significant decrease of Kₜ for increasing Hₜ (Figure 1a) which is in line with previous computational results [1]. Though we could show that Kₜ decreases with RBC velocity for Hₜ=2.2% (Figure 1b), we did not find sufficient statistical evidence to prove that Kₜ changes with RBC velocity in general. The two main limitations of this study are the low hematocrit and the square cross-section of the channel. We anticipate that future in vitro experiments will include circular channels and they will cover broader ranges of hematocrits and RBC velocities.

![Figure 1](image_url)

**Figure 1:** (a) Kₜ in function of tube hematocrits; (b) Kₜ in function of velocity for two different tube hematocrits (* indicates statistical significance, p < 0.05)

**References**

Dispersion and transit time of red blood cells in capillaries.

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Blood is a concentrated suspension made mainly of plasma, a Newtonian fluid, and of red blood cells (RBCs), which are deformable. This suspension flows in a complex and expanded ramified network. In this network, different coupled phenomena produce a complex flow. An old in-vivo study \cite{1} has highlighted the high dispersion of the time needed by RBCs to pass through an organ; besides, an other important result of their study showcases that rigidified RBCs had a higher dispersion than healthy RBCs. The exchange of gas inside the organs being proportional to the transit time of this RBCs and knowing that some diseases increase the rigidity of RBCs, it appears to be important to have a better understanding of the physics behind this phenomenon. The transit time mostly depends on the velocity of the RBCs and the paths chosen inside the vascular network. The velocity depends on the position inside the channel: the more a RBC is close to the center of the channel the faster it is. The position of the RBC is the result of an equilibrium between a lateral migration from the wall induced by the deformability of the cell and a dispersion due to hydrodynamic interaction between RBCs \cite{2}. By measuring the elongation of a bolus of RBCs in a rectangular micro-channel, a dispersion in the transit time of the RBCs can already be observed. This elongation is proportional to the difference of velocity between the faster and the slower RBC. Since velocity of RBCs is position dependent we can then quantify the lateral migration and the hydrodynamic dispersion, which are poorly documented in the literature, although they contribute to the creation of the cell free layer near the walls. The existence of such a layer is the key factor to explain many features of blood microcirculation such as the Fahreus Lindquist effect (the dependency of the apparent viscosity on the vessel diameter) or the plasma skimming effect at bifurcations. Preliminary results for dispersion in honeycomb channel will be presented and discussed too.

\textbf{Figure 1:} The red zones represent the zones where we can find RBCs bolus. The top sketch shows the plane (x,z) which is observed by the microscope. In this plane, the channel is large and the velocity field is flat. The bottom sketch shows the orthogonal plane (x,y) where the channel is narrow. In this plane the walls and the shear stress created by the velocity gradient induce cell migration in opposition to the hydrodynamic dispersion due to other cells. The evolution of the initial pulse length $L_0$ can be related to the structure evolution of the pulse in the (x,y) plane.

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Blood flow in biomimetics micro-channels

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Microvasculatures-on-a-chip, i.e. in vitro models that mimic important features of microvessel networks, have gained increasing interest in recent years. Such devices have allowed investigating pathophysiological situations involving abnormal biophysical interactions between blood cells and vessel walls. Still, a central question remains regarding the presence, in such biomimetic systems, of the endothelial glycocalyx. The latter is a glycosaminoglycans-rich surface layer exposed to blood flow, which plays a crucial role in regulating the interactions between circulating cells and the endothelium. Here, we use confocal microscopy to characterize the layer expressed by endothelial cells cultured in microfluidic channels. We show that, under our culture conditions, endothelial cells form a confluent layer on all the walls of the circuit and display a glycocalyx that fully lines the lumen of the microchannels. Moreover, the thickness of this surface layer is found to be on the order of 600 nm, which compares well with measurements performed ex or in vivo on microcapillaries. Furthermore, we investigate how the presence of endothelial cells in the microchannels affects their hydrodynamic resistance, the near-wall motion of red blood cells and how enzymatic removal of the glycocalyx affects circulating cell / endothelial cell interactions. Our study thus provides an important insight into the physiological relevance of in vitro microvasculatures.

References
Velocity, haematocrit and aggregation characteristics of stiffened RBC suspension flows

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Red blood cells (RBCs) are the most abundant cells in blood and their role on hemodynamics is significant. Their remarkable deformability allows them to pass through microvessels with diameters smaller than their size in order to deliver oxygen and nutrients. Diseases such as diabetes, sickle cell anemia and malaria, affect the morphology and mechanical properties of RBCs altering their haemodynamic behavior and leading to microvascular complications. However, the exact role of RBC deformability on microscale blood flow has not been established. In the present study we extend our previous work on healthy RBC flows in bifurcating microchannels [1,2] to quantify the effects of impaired RBC deformability on the velocity and haematocrit distributions in microscale blood flows.

RBCs were obtained from a healthy donor according to an approved ethics protocol (Ref:10/H0804/21) and various levels of membrane stiffening were introduced by glutaraldehyde (GA) at concentrations of 0.04% and 0.08% v/v in order to obtain up to three times reduction in RBC deformability [3]. RBC solutions were perfused through straight microchannels with a square cross sectional area of 50 x 50 μm². An unfixed RBC sample was also prepared and used as control as well as a 7-days aged set of healthy samples stored at 4 °C. Two different haematocrit levels (10% and 25%) were studied. Aggregation was induced using Dextran 1500-2800 at a concentration of 8 g/L. The flow was analyzed using a bright-field μPIV technique described elsewhere [1]. The velocity and haematocrit distributions across the channel were determined for flowrates ranging from 0.5 μl/min to 10 μl/min. The bluntness of the measured profiles was utilised to illustrate the effect of altered RBC deformability on blood flow dynamics.

The bluntness of the velocity profiles was found to progressively decrease with RBC stiffening in all cases studied. Furthermore, RBC stiffening reduced the extent of aggregation. As a result the velocity profiles of stiffened RBCs exhibited the same behaviour with and without aggregation.

References
Blood microstructure and viscosity in bifurcating microfluidic flows

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Abstract
The microstructure of blood is dominated by the presence of the red blood cell (RBC) and its tendency to aggregate under low shearing conditions. The effects of altered microstructural characteristics on blood flow are more pronounced in the microscale, where it has been shown that the bluntness and skewness of the velocity profile are affected by the phenomenon of RBC aggregation [1]. In addition, the local RBC concentration, the cell depleted layer and the distribution of the RBC aggregates in bifurcating geometries are influenced by the aggregation phenomenon [2,3]. RBC aggregation also affects the mechanical properties of blood, however, little information exists in the literature regarding local viscosity characteristics in the microscale. In the present work viscosity is characterised at the local scale using constitutive equations of blood from earlier work for flows in a T-type bifurcating microchannel. Viscosity fields are derived for various flow distributions in the channel, and the location of maximum viscosity magnitude is obtained. The viscosity does not appear significantly elevated in the branches of lower flow rate, and the maximum magnitude appears in the vicinity of the junction, towards the side of the outlet branch with the higher flow rate.

References
Shapes and positions of red blood cells in capillary flow

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A phase diagram of red blood cells (RBCs) flowing in a rectangular microcapillary is presented, i.e. we identify and characterize the shapes in dependence of flow parameters, such as flow rate and channel geometry. We hereby focus on experimental data, obtained by tracking of single RBCs flowing through a microfluidic device. Additionally, we provide a comparison of these results to 3-D simulations based on the boundary-integral technique. Among the identified single cells, we distinguish three shapes: A croissant-like shape, slippers and others.

Both numerical and experimental data show the appearance of croissants in a specific flow regime, whereas above a critical flow rate, these croissants become absent and slippers dominate. Moreover, the croissants show a migration towards the channel center, contrasting an off-centered (bimodal) distribution of slippers.

Figure: Examples of croissants (left), slippers (center), and others (right).

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Rheology of concentrated red blood cell suspensions and cell dynamics in flow

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Erythrocytes (or red blood cells – RBCs) under shear flow exhibit various dynamical behaviours [1-3]. We studied experimentally the viscoelasticity of concentrated red blood cell suspensions with cell volume fractions above 0.1 [2]. For concentrated suspensions, the individual cell dynamics in flow is altered by the hydrodynamic interactions between cells. The results lead to the conclusion that rheology of concentrated suspensions is also sensitive to the cell dynamics similarly to the dilute limit [4]. At small shear rates (<10 s⁻¹) the effective viscosity of RBC suspensions is measured to depend on the prehistory of sample shearing. At a given viscosity of the external medium the higher RBC volume fraction is, the stronger this dependence is obtained. The hysteresis effect is found under moderate shear stresses, at which swinging of RBC is expected [5]. Rheological measurements on concentrated RBC suspensions accounting for the individual dynamics of cells are indicative of cytoskeleton-related phenomena and alterations of the cellular elasticity.

References


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Flow vortices induced by red blood cells

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Red blood cells (RBCs) are very deformable objects travelling inside vessels in our body. During their journey between the arteries and capillaries, they undergo different flow and shear stresses affecting their shape. In our experiments, we mimic these small capillaries by using microchannels of 10 micrometers in width. RBCs were drawn from a single donor and resuspended in a solution of PBS in presence of poly ethylene glycol (PEG) nanoparticles with a diameter of 250 nm. In this work, we monitor the trajectories of these nanoparticles in the comoving frame of the RBCs by using a high speed camera and a particle tracking algorithm. The particles' displacements provide us with information on the flow induced by the passage of RBCs in the capillaries such as relative velocity for several mean flow velocities. Vortices have been found in the vicinity of RBCs and characterized between two cells.
Effects of red blood cell aggregation on microparticle margination in human blood

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Margination is the migration of particles in a channel towards the outer walls of the channel. In blood microcirculation, studying the margination of microparticles is important to understand platelet migration and the kinetics of drug delivery. Many new topics in drug delivery research examine the slow release of drugs through microparticles, such as micelles. The margination of such drug carriers is related to tissue absorption and, consequently, to the efficiency of drug delivery. We hypothesized that the intensity of red blood cell (RBC) aggregation will change the level of margination in a cylindrical channel. RBC aggregation is the reversible process of RBCs clumping together over time, under low fluid shear rate. A higher level of aggregation means that this clumping occurs more quickly.

The goal of this study was to design an experiment that quantifies the level margination of microparticles and the effect that RBC aggregation has on margination, in a controlled in vitro environment. Fluorescent microparticles were added to human blood preparations. The aggregation properties of the blood preparation were modulated by the addition of a macromolecule (dextran 500). The blood preparations were injected into PDMS microfluidic devices that were modified to have circular channels in order to better mimic the geometry of physiological microcirculation.

Circular micro-channels were successfully designed that worked to capture the marginating microparticles. It was found the level of margination of the microparticles increased with an increase of aggregation of the RBCs. This increase in margination was especially sensitive to aggregation in the range of physiological aggregation levels of whole blood, suggesting that aggregation may play a role in margination in vivo, in particular for diseases associated with hyperaggregation such as diabetes.
A multidisciplinary/multimodal approach to study the prothrombotic effect of shear rate gradient

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Fibrinogen plays a major role in both the primary and secondary hemostasis. It is highly expressed in the plasma. Fibrinogen promotes platelet-platelet interaction at site of vessel injury, supporting thereby the formation of a hemostatic plug, which stops blood loss. In the secondary hemostasis the biological function of fibrinogen is mainly devoted to the furniture of fibrin (the insoluble form of the protein) which occurs after its cleavage by thrombin, a mechanism known as the common pathway of the coagulation cascade. This process stabilizes the formed clot. This process can also take place in a diseased artery presenting a thickening of the vessel due to atherosclerotic plaque formation. Rupture of such a plaque precipitates platelets aggregation leading to the formation of a thrombus than can become occlusive. This results in life threatening ischemic pathologies such as stroke or myocardial infarction.

Surprisingly, even though fibrinogen is central for primary and secondary hemostasis, it is considered to be a weak adhesive protein for platelets when it is immobilized on a surface. In contrast, VWF (von Willebrand factor) meditates platelet recruitment at elevated wall shear rate after adsorption on various immobilized subendothelial proteins including collagen fibers. To date the role of fibrinogen and VWF under pathological blood flow conditions as those found in diseased arteries remains poorly understood.

In vitro flow chambers are powerful tools to study the function of adhesive proteins [1][2]. In the present work we conducted several parallel explorations based on a microfluidic flow chamber: i) Flow based assays to measure thrombus formation, ii) Experimental flow characterizations to assess experimentally the flow in the different regions of the model which mimics a diseased artery (stenosed vessel), iii) Numerical flow and molecular dynamics simulations in order to access various unmeasurable data.

The results from this multidisciplinary approach tends to confirm the implication of VWF unfolding and vWF fiber like formation generated by shear rate gradient (SRG) in an exponential platelet aggregation process on fibrinogen.

References

Blood is a complex multiphasic fluid for which optical and rheological properties are mainly governed by the interactions between Red blood cells (RBCs) and plasma. While plasma is a simple Newtonian fluid, RBCs are small microscopic deformable vesicles made of a thin membrane enclosing a hemoglobin solution. Because of the negligible thickness of the RBC's membrane their optical parameters correspond essentially to those of the internal hemoglobin solution and to multiscattering effects due to high concentration of RBCs. Their shape, concentration and deformability play a major role in the behavior of blood under flow; in particular shearing of RBCs can result in aggregate formation (low shear), orientation in the flow direction (medium shear) or deformation (high shear) [1]. Due to various technical limitations but mainly to the considerable visible light attenuation in undiluted blood, in vitro blood flow studies have often been limited to the use of transparent blood mimicking fluids or to cases of low hematocrit or flow conditions, most not physio-pathologically representative. To overcome these problems and limitations, transparent suspensions can be prepared by using RBC ghosts (i.e. hemoglobin free RBCs) [2] and can be put in suspension to reproduce the rheological behavior of normal blood at high hematocrit.

In the present work RBC Ghosts (RBC-Gh) are prepared by a bulk hypotonic extraction of hemoglobin from porcine blood, suspended in a transparent fluid. The resulting transparent 45% suspension shows a rheological behavior in close relationship with the behavior of human blood at medium to high shear rates and enables microPIV measurements of the flow fields in 200μm diameter microchannels under realistic flow conditions.

**References**


Shape and position of moving red blood cells in microchannels

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We investigate experimentally the shape and positions of red blood cells as function of time, driven by an Poiseuille flow. Microfluidic channels with various width was used. The cells were observed by an optical microscope with a motorized stage. We could establish a real-time program to detect the cell position in the current observation image and a feed-back algorithm to follow the moving cell in the channel with the help of the motorized stage. Therefor, we are able to follow moving cells over distances of several cm at a speeds up to 2 mm/s, with a recording frame rate up to 100 Hz, see Figure.

An automatic procedure records the cell movements in a loop. We can classify the temporal development of the cell shape as function of the time/distance from the entry. Additionally, if two consecutive cell in the observation window, the cell distance in the co-moving frame is determined. We use our data to compare the measurements with predictions from a theoretical treatments [1].

References

Role of human red blood cell membrane stiffness on time of passage through narrow straight channels

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The cell membrane stiffness (CMS) of Red Blood Cells (RBCs) plays an extremely important role in the process of passage of these cells through capillaries in the human vasculature that have diameters smaller than the RBC diameter. Alteration in the CMS of RBCs is associated with a number of medical conditions which can be expected to also result in an altered micro circulation of these cells. Here we report on the effect of altered RBC CMS on the passage time through narrow straight channels.

We have developed a controlled way of tuning the CMS of RBCs that enables us to conduct studies on the effects of altered RBC CMS without actually using diseased cells. This is done by adding Bovine Serum Albumin (BSA) to the phosphate buffered saline (PBS). Two different methods using optical tweezers (OT) have been used to study the effect of BSA treatment on the CMS of RBC. The first method involves the measurement of the reorientation time of RBCs entering the Optical trap as a function of laser power and using an analysis protocol described in [1] to gauge the change in CMS. This method establishes that RBCs suspended in PBS with 0.5 mg/ml of BSA behave like normal RBCs. On the other hand, RBCs treated with BSA concentrations below 0.5 mg/ml show a reorientation response commensurate with a lowering of CMS, while those of RBCs treated with BSA concentrations greater than 0.5 mg/ml indicate enhanced CMS. The other method involves recording Brownian fluctuations of trapped and reoriented RBCs using a Quadrant Photo Diode (QPD) and extracting a corner frequency ($f_c$) from a power spectral density analysis of the recorded fluctuations. We find that though an identical laser power is used throughout, $f_c$ reduces as the concentration of BSA is increased. Assuming that the viscous drag coefficient remains unchanged with BSA concentration, the apparent trap stiffness could be said to reduce with increase in BSA concentration which would be commensurate with an increase in RBC CMS. We find an almost linear relation between the factor serving as a marker for CMS from the reorientation experiments in the OT and $f_c$ extracted from a PSD analysis.

We use these BSA treated RBCs to study the effect of RBC CMS on the time required for RBC deformation to enable passage through a narrow channel. To do this, we use cyclopore filter papers which are comprised of uniform straight channels with identical pores sizes comparable to blood capillary diameters. From a comparison of the number of RBCs that filter through per unit time and per unit volume of suspension fluid to the corresponding number at the input stage, we extract the time of deformation. We compare these times with those computed using a simple theoretical model where an axisymmetric shape of the RBC on deformation and a constraint of constant surface area during the process of deformation is assumed. We find a strong linear correlation between the experimentally measured and theoretically predicted RBC deformation times.

References
Influence of erythrocyte aggregation on radial migration of platelet-sized spherical particles in shear flow

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Blood platelets when activated are involved in the mechanisms of hemostasis and thrombosis, and their migration toward injured vascular endothelium necessitates interaction with red blood cells (RBCs). Rheology co-factors such as a high hematocrit and a high shear rate are known to promote platelet mass transport towards the vessel wall. Hemodynamic conditions promoting RBC aggregation may also favor platelet migration, particularly in the venous system at low shear rates. The aim of this study was to confirm experimentally the impact of RBC aggregation on platelet-sized micro particle migration in a Couette flow apparatus. Biotin coated micro particles were mixed with saline or blood with different aggregation tendencies, at two shear rates of 2 and 10 s⁻¹ and three hematocrits ranging from 20 to 60%. Streptavidin membranes were respectively positioned on the Couette static and rotating cylinders upon which the number of adhered fluorescent particles was quantified. The platelet-sized particle adhesion on both walls was progressively enhanced by increasing the hematocrit (p < 0.001), reducing the shear rate (p < 0.001), and rising the aggregation of RBCs (p < 0.001). Particle count was minimum on the stationary cylinder when suspended in saline at 2 s⁻¹ (57 ± 33), and maximum on the rotating cylinder at 60% hematocrit, 2 s⁻¹ and the maximum dextran-induced RBC aggregation (2840 ± 152). This fundamental study is confirming recent hypotheses on the role of RBC aggregation on venous thrombosis, and may guide molecular imaging protocols requiring injecting active labeled micro particles in the venous flow system to probe human diseases.
Competition between aggregation and break up of Red Blood Cells: Depletion force due to filamentous viruses vs shear flow

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Blood is a shear thinning fluid with a complex response that strongly depends on the Red Blood Cells (RBC) ability to form aggregates in the form of stacks, called rouleaux. [1] Both, depletion and bridging between RBCs have long been believed to play a role in rouleaux formation, mediated by the presence of macromolecules such as fibrinogen in blood plasma. However, despite numerous investigations, microscopic understanding of the formation and break up of RBC aggregates has not been fully elucidated. [2]

In order to distinguish the mechanisms behind RBCs aggregation we employ a depletant agent with very long ranged interaction force, namely the filamentous fd bacteriophage. [3] These colloidal rod-like particles have high length-to-diameter ratio and carry the same charge as the cells. Fine-tuning of the depletion force is achieved through mixtures of RBCs in PBS at different concentrations of rods. We study the breakup of aggregates during shear flow to quantify the interaction between the cells, combining a home-build counter-rotating cone-plate shear cell with an ultra-fast confocal microscope enabling visualization of the structures while shearing.

We present the non-equilibrium phase diagram of the shear rate versus the concentration of the depletant, showing regions for different flow response and stages of separation of the RBCs doublets.

References


Torque-free elastic moduli
in spring network model of red blood cell

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In the absence of external stimuli, computational model of elastic cell should exhibit neither spontaneous movement nor rotation, i.e. any elastic forces and resulting torques need to balance out. The model of red blood cells in fluid described in [1] includes five force-based elastic moduli: stretching, bending, local and global area conservation and volume conservation. In [2], we have discussed the force- and torque-free conditions for the area forces.

Here we apply similar approach to bending modulus, which was originally implemented as $F_b(ABC) = \beta_b n_{ABC}$, where, $\beta_b = k_b (\Delta \theta / \theta_0)$, $k_b$ is the bending coefficient, $\theta_0$ is the resting angle between two triangles that have common edge $AB$, $\Delta \theta = \theta - \theta_0$ is the deviation from this angle and $n_{ABC}$ is the outer unit normal vector to the triangle $ABC$. This force is applied to the vertex $C$ and half of this force with the opposite direction is applied to the vertices $A$ and $B$. Analogous force is applied to point $D$ of the adjacent triangle $ABD$.

This definition of bending modulus is force-free but with equal distribution between points $A$ and $B$, it is not torque-free. To make it torque-free, we first parametrized the forces $F_b(C)$ and $F_b(D)$. Next we parametrized the portions of $-F_b(C)$ and $-F_b(D)$ assigned to points $A$ and $B$. Zero torque calculation reveals the values of parameters. With careful computation it can be verified that up to a multiplicative factor $\beta_b$, these bending forces correspond to those described in [3, 4], which were obtained by different approach - by differentiating the bending energy.

Similar analysis of volume force shows that $F_v(A) = -k_v \beta_v S_{ABC} n_{ABC} / 9$ is both force- and torque-free. Here $S_{ABC}$ is the area of triangle $ABC$. This force is different from the volume force $F_v^E(A) = -k_v \beta_v (B \times C) / 6$ derived from volume energy, which does not act solely in the direction perpendicular to the given triangle. This means that the tangential component of $F_v^E$ may interfere with the action of the global area conservation force acting in plane: one of them trying to decrease the size of the mesh triangle and the other to increase it. Such consequences can be avoided by using $F_v$.

References

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Microparticle Margination in complex geometries

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Complex geometries frequently occur in blood circulation and play an important role for drug delivery.
First, consider a mixed suspension of red blood cells and microparticles flowing through a cylindrical channel with a constriction mimicking a stenosed blood vessel (Fig. 1 left). Our three-dimensional Lattice-Boltzmann simulations show that the red blood cells are depleted right ahead and after the stenosis. For the red blood cells the axial concentration profile is very similar to that of isolated tracer particles flowing along the central axis. Thus, the depletion of red blood cells is explained by the acceleration while entering the stenosis. Most importantly, however, we find that the stiff microparticles exhibit the opposite behavior. Arriving on a marginated position near the channel wall, they can pass through the constriction only if they find a suitable gap to dip into the dense plug of red blood cells occupying the channel center. Hence, clustering of microparticles ahead of the stenosis can be explained by the interplay of stenosis and margination [1].

Second, we investigate the margination of microparticles in a red blood cell suspension flowing either through a branching channel or through channels, which merge after a certain distance (Fig. 1 right). The geometry is modeled with the help of inflow and outflow conditions at the boundaries of the system in flow direction. Flowing towards a bifurcation into two channels both red blood cells and microparticles located at the mid-plane are slightly trapped at the apex of the bifurcation. At the end of the main channel the cell-free layer, the near-wall region with zero red blood cell concentration, decreases and we obtain an asymmetrical cell-free layer inside the branches. Microparticles arriving near the upper and lower boundary of the system, respectively, show a peak in concentration directly at the beginning of the branches. Corresponding changes in cell-free layer and microparticle concentration occur right in front of the merging, as well.

Fig. 1: A suspension of red blood cells and microparticles flowing either through a stenosed channel (left), a bifurcating channel (middle) or two merging channels (right).

References
The effects of pulsatility in 2D cell resolved blood flow simulations of curved vessels with aneurysms

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Pulsatility can influence the transport of red blood cells and platelets. To study this effect in a curved vessel with an aneurysm we use a validated [1] two dimensional lattice-Boltzman model for the blood plasma with a discrete element method for both red blood cells and platelets. The flow was driven by a time varying body force applied to a periodic pre-intlet domain which is coupled to the curved vessel with an aneurysm. The accuracy of the inlet boundary condition was first checked by comparing the velocity profile in a straight pipe from a sinusoidal varying pressure curve to the expected profile predicted by the Wormersley solution. Agreement is good between the computed and predicted flow profiles with errors of approximately 0.1% after 10 heart cycles. Flow velocities and vessel diameters were then matched with measurements of cerebral perforating arteries [2], and the flow was driven by a synthetic heartbeat curve typical for such vessel sizes. The effect of pulsatility on the aggregation of both red blood cells and platelets in the aneurysm was probed by varying diameter to neck aspect ratios (1.0, 2.0, 3.0 and 4.0) of the aneurysm. The local shear rates in the suspending fluid plasma can help indicate regions of a shear rate threshold where thrombus formation could occur [3]. Combining local stresses in the fluid along residence times of the cells in the aneurysm could provide insight for thrombus forming regions. These simplified 2D simulations can help shed light on cases to be studied in greater detail with 3D models.

Fig. 1: Plasma fluid shear stresses (left) and residence time of red blood cells and platelets (right) are shown for a simple 2D curved vessel with an aneurysm with a diameter to neck aspect ratio of 2.0.

References
In-silico investigation of the effect of cytoplasm viscosity on blood transport mechanics

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Blood is a dense cellular suspension with unique transport properties, which gives rise to several complex phenomena, such as the margination of platelets, the Fåhræus effect, or the appearance of a cell-free layer. The knowledge of the emergent complex rheology is gaining importance due to the spreading application of micro-medical devices in numerous clinical applications during the last years. In the current work, we investigate the link between these effects and cytoplasm viscosity through cell-cell collision mechanics using the Hemocell framework [1] which is capable of simulating accurate cellular flows (e.g. see Fig. 1). The plasma is resolved by applying the lattice Boltzmann method, while the cellular membranes are implemented using a discrete element method (DEM) coupled to the fluid as immersed boundary method surfaces. The mechanical behaviour of the cells is expressed by a set of forces using this DEM membrane structure. The single-cell mechanical responses were validated using optical tweezer stretching and ectacytometry shearing measurements available in the literature.

Red blood cells (RBCs) have viscoelastic responses to mechanical deformations [2], which can influence the aforementioned transport properties of blood. The viscous part of the response is controlled mainly by the density of the cytoplasm, which in turn depends on several variables, for instance the hemoglobin concentration. This viscosity yields an additional term in the force exchange of the colliding cells depending also on the velocity of the collision, thus it modifies the local stiffness of the cells. The relative stiffness of the colliding cells has impact on their trajectory which influences several transport properties, such as the margination [3]. We investigate these collision trajectories of the three most numerous cell types: RBCs, platelets, white blood cells and their dependence on cytoplasm viscosity and flow shear rate.

Figure 1: Flow of RBCs (red) and platelets (yellow) in a straight vessel section (D=128 µm, H=45%) computed with Hemocell.

References
A computational method is presented for generating synthetic, random 3D capillary networks which match the topological, geometrical and functional properties of the cerebral microcirculation. These networks, which can be generated in volumes larger than can currently be extracted by high-resolution imaging, can then be coupled to lower-resolution data sets of whole-brain vasculature to model blood flow and mass transport, and to validate equivalent continuum/hybrid models. Another motivation is to reveal the dominant structural features of cerebral capillary networks, which can then be tuned to model different brain regions or pathological states such as Alzheimer’s disease. Previous works [1, 2] lacked physiological basis, and although resulting networks conformed to expected global morphometric properties, were not subjected to thorough topological or functional analysis.

In contrast, our approach is based on the physiological assumption that the maximum separation of tissue cells from the nearest capillary is limited by the diffusion distance of oxygen [3]. Previously, synthetic, space-filling 2D networks were constructed by placing one point randomly in each cell of an \( n \times n \) grid; from this set of points, Voronoi diagrams were extracted with the edges producing a 2D network with mainly three capillaries per vertex, a characteristic feature of cerebral capillary networks. Here, we extend this approach to 3D.

In 3D, Voronoi diagrams produce polyhedrons with many capillaries per vertex. To derive a network with only bifurcations, clusters of vertices were systematically merged and capillaries then removed randomly. Geometrical metrics such as the mean/S.D. of lengths and edge/length/vertex densities were compared to those of capillary regions extracted from mouse cerebral anatomical data sets [5, 6]. Capillary loops were studied to measure the interconnected network topology, while the distribution of extravascular distances allowed comparison of the spatial arrangement of capillaries. Finally, hemodynamic properties were captured through the network permeability. Overall, synthetic networks showed excellent agreement with the anatomical data.

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References

Demargination and Mechanical Dissociation of Platelet Aggregates in Blood Stream

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Platelet aggregation in blood stream is necessary in blood clotting. The emergence of plasma proteins von Willebrand factors (VWFs) and their stretching in high shear rates is a key phenomenon in the initiation of platelet aggregates. VWFs are very large polymers of dimeric protein units [1] which are collapsed in low shear rates and stretch once the shear rate surpasses a critical value [2]. This critical stretching behavior leads to the critical formation of platelet-VWF aggregates in shear rates beyond the critical shear rate of VWF stretching. Interestingly, such aggregates dissolve reversibly when the shear rate gets under the critical value [3].

In blood stream, red blood cells (RBCs) are hydrodynamically forced to the center of the vessel, leaving a RBC free layer (RBC-FL) close to the walls. Their concentration at the center of the flow pushes other components such as platelets and large VWF polymers to the RBC-FL [4]. The margination of platelets and VWFs increases the chance of aggregation in RBC-FL. Particularly, the shear rate in RBC-FL is an order of magnitude higher than the shear rate at the bulk of the flow and makes the aggregation possible. Based on the mesoscale hydrodynamic simulations of blood flow in microchannels, it is found that the aggregates actually form in RBC-FL and grow until they are lifted up to the bulk flow. Their demargination to the zone of low shear rates results in their dissociation and remargination of platelets and VWFs. This process is dependent on the VWF properties and their interaction with platelet receptors that a mutation in VWF or its protease can result in irreversible fatal thrombosis or deficiency in blood clotting.

References
Upscaling mass transfer in brain capillary networks

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Various imaging techniques are now capable measuring spatio-temporal concentration fields of various endogenous or exogenous tracers in the brain. For example, the concentration of radio-labeled water injected transiently in the vascular system can be measured by Positron Emission Tomography (PET), while the presence of paramagnetic deoxy-hemoglobin induces contrast in functional Magnetic Resonance Imaging (fMRI). The resolution of PET is about \((10 \text{ mm})^3\) while fMRI can achieve resolutions down to \(1 \text{ mm}^3\). This is much coarser than the diameters of most arterioles and venules, which are typically below \(100 \mu\text{m}\), and, of course, of capillaries, whose diameters are tenfold smaller.

This implies that methods to deduce the regional blood flow rate out of these large-scale concentration fields must rely on upscaled models which take into account the micro-structure of the vascular system. Consequently, capturing the strong link between micro-vascular structure and mass transfer at mesoscopic scales is a major hurdle in the challenge to better understand and exploit the information obtained by these imaging techniques, as discussed in [1].

It has been shown in [3] that at the capillary level, the vessels exhibit a space filling mesh-like structure, for which a Representative Elementary Volume (REV) can be extracted. This property allows for the use of the Volume Averaging Technique on the advection-diffusion equations at this particular scale. This method has been developed for upsampling mass transfer in heterogeneous porous media [2]. It requires solving closure equations on a REV to deduce effective coefficients representative of the micro-structure. The REV is a 3D network of capillaries with diameters ranging from 1 to 10 \(\mu\text{m}\) embedded in tissue. Its typical volume is about \((150\text{ to }300 \mu\text{m})^3\). Being able to solve closure equations on several geometries taking into account the presence of individual vessels is a computational challenge.

We developed a numerical framework to solve partial differential equations on anatomically accurate vascular networks using the high performance finite element library Feel++ [4]. This framework is used to solve the closure equations on a VER needed to get the effective coefficients from the homogenization method as well as to perform direct simulations of mass transfers to check the homogenization procedure. In the first part of the presentation we will present this framework, with focus on, the way to obtain a mesh adapted to vascular networks. Then, we will detail the procedure to obtain effective coefficients in the capillary network. Finally, we will show that the homogenized and the direct approach are in good agreement.

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References

In silico investigation of rheological characteristics of a red blood cell suspension in a simple shear flow

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Numerical experiments on the rheology of suspensions of aggregation-free and aggregate red blood cells (RBCs) in a simple shear flow are performed by the lattice Boltzmann method coupled with a coarse-grained membrane model composed of a spring network. We first match the RBC elasticity and aim to capture the whole blood viscosity dependence on the shear rate and RBC volume fraction. Adhesive forces among RBCs are taken into account to simulate rouleaux formation and to study consequences of RBC aggregation.

Our model successfully captures the shear-thinning behavior of the suspension as the shear rate increases, in qualitative agreement with experimental observations. We find that the suspension viscosity increases nonlinearly with the particle volume fraction. As the shear rate increases, the normal components of particle stress in the shear and vorticity directions increase, and the particle pressure decreases. These are attributed to flow-induced RBC alignment and weaker inter-particle interaction at larger shear rates. The relative viscosity, particle pressure, and normal stress differences are investigated for a range of hematocrits and shear rates.
Hydrodynamic crystals in weakly confined shear flow

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We have shown recently experimentally and by numerical simulations that dilute dispersions of red blood cells or other soft particles can exhibit spontaneous ordering when subject to a shear flow between two walls. The ordering is attributed to a complex interplay between four effects of the hydrodynamic origin: (1) wall-induced migration of particles away from the walls, (2) long-range hydrodynamic attraction, (3) short-range hydrodynamic interactions that push the particles in the direction orthogonal to the walls and (4) advection by the shear flow.

We base our model on asymptotic expansions of the hydrodynamic interactions between the particles in confined geometry, assuming the size of the particles to be small compared to the interwall distance. Our model shows that after the particles migrate towards the midplane between the walls due to the wall-induced lift, the order appears as a competition of the long-range attraction and the short-range repulsion. The repulsion in a pair of particles results from advection by the shear flow because the particles end up having opposite displacements from the midplane between two walls. The displacement appears as a competition of short-range transverse repulsion between the particles and the wall-induced migration.

Our model shows that stable stationary configurations are possible for two or more soft particles in confined shear flow. Further, we analyze the excitation spectra and the stability of periodic one-dimensional arrays and two-dimensional lattices of soft particles.
Red blood cells distribution in microvascular networks: a model derived from experiments

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Blood flow and red blood cells (RBC) distribution across the microvasculature play an essential role in delivering oxygen and nutrient to tissues. In living organisms, microvascular networks form large and complex structures with tens of thousands of vessels [1] in which blood and RBC flows exhibit highly heterogeneous repartitions. This behaviour is challenging to interpret without theoretical and numerical frameworks derived from controlled experiments.

Here, we first focus on modeling blood flow and RBC distributions across simple artificial network geometries (square, honeycomb) with different diameters (5µm, 10µm, and 20µm) in order to enable comparisons with in vitro measurements where human RBCs flow in the same networks. Then, we consider large anatomical networks, and compare simulation results to in vivo experiments.

Due to the size of these networks, each RBC cannot be described individually. We consider the blood as a monophasic, non-Newtonian fluid whose rheology strongly depends on vessel diameter and discharge hematocrit. Further, the repartition of RBCs between branches at bifurcation is unequal and non-linear. This phase separation effect has been approximated by empirical laws such as [2] in simple diverging bifurcation. We solve this coupled, highly non-linear problem with an algorithm based on an iterative solver inspired by [3].

We validate this approach by direct comparison with in vitro network measurements of hematocrit and RBC velocity. While we find very good agreement for artificial networks with small diameters (5µm), significant discrepancies arise for larger ones (> 10µm). In the latter, RBCs are still influenced by the splitting at the previous bifurcation when entering the next one. This results in an asymmetry in hematocrit profile. Taking this asymmetry into account in the model reduces the prediction error by more than a half.

Recent work [4] points out that the above coupling between hydrodynamics and hemodynamics may lead to several stationary solutions due to non-linear effects. We investigate these multiple equilibria in simple geometries and show that the differences between them is, on average, below experimental error. The model has been extended to solve the blood flow in large anatomical networks [1] and the first comparisons with in vivo measurements in the mice cerebral cortex [5] shows very good agreement with simulations.

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References
Effect of red blood cells on wall shear stress and flow properties using lattice Boltzmann-immersed boundary methods

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The effect of red blood cells and the undulation of the endothelium on the shear stress and flow properties in the microvasculature is here studied in detail using the lattice Boltzmann-immersed boundary method (LB-IBM). The results demonstrate a significant effect of both the undulation of the endothelium and red blood cells on wall shear stress. The resulting fluctuations in wall shear stress greatly exceed the mean values, emphasizing the importance of the particulate nature of blood as well as a more realistic description of vessel wall geometry in the study of hemodynamic forces. Our results also reveal that morphological alterations of red blood cells, as occur in certain pathologies, can significantly affect the values of wall shear stress observed. We will also look at how flow properties such as apparent viscosity are also greatly altered when suspensions of cells are considered in wavy-walled channels.

Figure 1 – Single cell exhibiting parachute morphology in a wavy-walled channel; Bottom : Plots of the ratio of the wall shear stress with the cell to the Poiseuille solution (effect of the wavy wall and the effect of the cell on the wall shear stress are both captured) and plot of the ratio of the wall shear stress normalized to the wavy wall solution (effect of cell on wavy wall is captured)
A computational study on the effect of plasma skimming on vascular development

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In recent work, we have pioneered the developed computational models of retinal blood flow for the study of vascular development [1, 2, 3]. In these works, blood was modelled as a shear-thinning homogeneous fluid rather than a particle suspension. This simplification, often required for computational tractability reasons, is appropriate for the study of haemodynamics in vessels with diameters larger than a few hundred micrometres. However, it fails to capture important rheological properties when applied to the simulation of blood flow in capillaries.

In the current work, we successfully implemented a model of deformable particles (red blood cells, RBC) [4] into the parallel open source HemeLB Computational Fluid Dynamics solver (http://ccs.chem.ucl.ac.uk/hemelb). We present example simulations in realistic developmental vascular networks and study the differences in the wall shear stress (WSS) predicted by a homogenous shear-thinning rheology model and the new particle suspension rheology model. Furthermore, we characterise how RBC deformability influences haematocrit partition at network bifurcations and how this affects the WSS distribution across the network. Based on our results, we hypothesise that plasma skimming enhances the WSS gradients known to modulate developmental vascular remodelling [2].

Figure 1: a) ICAM2 mask (blood vessel luminal marker) of a 6-day-old mouse retinal vascular plexus (a popular animal model for the study of vascular development) [1], b) RBC distribution at 20% inlet haematocrit and inlet capillary number of approximately 0.05, c) WSS traces at two points of interest (indicated by the origin of the red arrows).

References
Fully implicit Finite Element methodology for the modeling of biomembranes and red blood cells.

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This framework is concerned with the numerical modeling of the dynamics of individual red blood cells (RBCs) in a surrounding incompressible Newtonian fluid. The study of the deformations of RBCs is a tremendously challenging topic in theoretical and computational modeling, inducing consequently a growing interest over the past decade. The RBC’s deformability is an intrinsic mechanical property determined by the bending energy of Canham, Helfrich and Evans, in which the main mode of the deformation is bending, and the cost in the bending energy is driven by the curvatures of the membrane. A bending force highly nonlinear with respect to the cell’s shape drives consequently the deformations of RBCs [1]. The cell’s movement is dictated by the interplay between the hydrodynamic forces and the membrane force. At the numerical level, fully explicit decoupling strategies are usually considered, yielding a severe stability condition for the temporal resolution [2,3]. In fact, that results in the restriction of the time step size which depends on both the mesh size and the bending rigidity coefficient.

In this work, we develop a fully implicit finite element methodology tailored for the modeling of RBCs immersed in a Newtonian fluid [4]. Both exact Newton-Raphson and quasi-Newton strategies are investigated and guarantee the second-order convergence behaviour. A banded level set variant allows to handle the singularity of the global matrix while featuring, in addition to the parallel implementation, an affordable computational cost. We address in detail the main features of the proposed method, and we report several experiments with the aim of illustrating its accuracy and efficiency. Comparative investigations with respect to the fully explicit scheme depict the stabilizing effect of the new method.

References


Blood at physiologic conditions is a dense suspension of cells, dominated in terms of its dynamics by red blood cells, they make up over approximately 40% of the blood volume, and they are the blood component principally responsible for its rheology. RBCs are made of a two dimensional fluid bilayer of phospholipids, having underneath a network of proteins conferring to them shear elasticity. Simplified systems, like vesicles (made of a pure bilayer of phospholipid) and capsules (made of an extensible polymer shell) are used as models for RBCs, both systems reproduce several features known for RBCs under flow, the general problem is to understand the movement of cells under different flows and geometries. The model used in our 2D simulation is the Giant Unilamellar Vesicle (GUV) model, the membrane curvature force will be calculated with the Helfrich elasticity theory. The numerical resolution will give us the vesicle shape as function of two characteristic numbers the Capillary number ($C_k$) and the Confinement ($C_n$) [1]. A large number of studies were devoted to finding the equilibrium shapes of RBCs in Poiseuille flow [2]. We focused our study in a particularly interesting shape which has a motion like flagella, and called "Snaking Shape".

References
Does a vesicle migrate to the center or to the periphery in a bounded shear flow?

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The lateral migration of a suspended vesicle (a model of red blood cells (RBCs)) in a bounded shear flow is investigated numerically at vanishing Reynolds number (the Stokes limit) using a boundary integral method. We explore the relevant dimensionless parameters to study the dynamics and rheology of a vesicle as a function of the viscosity contrast $\lambda = \eta_{in}/\eta_{out}$, where $\eta_{in}$, $\eta_{out}$ denote the inner and the outer viscosities. A vesicle is found to migrate to the centerline or to the wall depending on $\lambda$. We found that below a critical viscosity contrast $\lambda_c$, the vesicle is centered, and above $\lambda_c$, the vesicle can be either centered or off-center depending on initial condition. The equilibrium lateral position of the vesicle exhibits a saddle-node bifurcation as a function of the bifurcation parameter $\lambda$, which leads to a surprising acute decrease of the effective viscosity of the suspension at a critical value of viscosity contrast ($\lambda_c$). This study can be exploited in the problem of cell sorting out and can help understanding the intricate nature of the rheology of confined suspensions.

![Fig. 1: (Color online) Phase diagram showing regions where migration is towards the center (if initial position is within yellow area) or towards the wall (if initial position is within orange area). The saddle-node bifurcation occurs at $\lambda_c \simeq 16$ for a reduced area $\tau = 0.7$ where $\tau = (A/\pi)/(L/2\pi)^2$ where $A$ the enclosed area and $L$ is the vesicle perimeter. The channel center is at zero and the wall is at $-0.5$. $W$ is the channel width and $R_0$ is the effective radius of the vesicle.](image)
Toward whole brain simulations of blood flows in human cerebral microcirculation: hybrid modelling and high performance computing

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The human brain microcirculation presents a dual multiscale architecture. On the one hand, the arteriolar and venular trees (10-100\(\mu\)m in vessel diameter) supply the cortex with blood, which carries oxygen and nutrients, and drain the metabolic waste. On the other hand, the capillaries (1-10\(\mu\)m in diameter) constitute a space-filling network connecting the larger arteriolar and venular trees. An important role of these capillaries is to facilitate molecular exchanges between blood and the cerebral tissue, therefore supporting the neuronal metabolic demand. Aging or cerebral diseases may induce architectural modifications in human brain microvascular networks, such as capillary rarefaction, which in turn may affect neuronal activity.

Modelling is key in understanding such systems and the systemic impact of localized effects, such as capillary stalling occurring early in Alzheimer’s disease. In particular, network approaches, which model dynamics at the scale of individual vessels, have significantly advanced our understanding of blood flow, mass transfers and regulation mechanisms [1]. However, such methods are still intractable at clinically relevant scales, typically the whole cortex, primarily because of the computational cost associated with the huge number of vessels involved.

Here, we present a hybrid approach to modelling blood flow in the microcirculation by treating the capillary bed as a continuum [2] and the arteriolar and venular trees as a network. The continuum model [3] characterizes the flow at a scale much larger than the length of a capillary and can be solved using finite volume methods on a coarse grid, therefore significantly decreasing the computational cost. The arteriolar and venular trees, however, have a quasi-fractal structure, thus cannot be homogenized and must be treated as a network. To capture the strong pressure gradients that build up in the vicinity of coupling sites, we introduce an analytical approximation (inspired by [4]) of the local pressure fields. The resulting coupling model consists in a single linear system describing both the network and continuum.

Comparisons between the hybrid and full network approaches show very good agreement for simple configurations with one or two coupling points, as well as for realistic structures displaying more than 200 coupling points (local pressure errors < 6 \%). The hybrid approach further yields an important computational gain, with an acceleration of 360 compared to the network approach. The accuracy and the computational benefit of the hybrid approach for blood flow modelling opens the way to include additional levels of complexity in the future and ultimately to simulate mass transfers in the whole brain.

References

Different dynamics of red blood cells define their trajectory in deterministic lateral displacement arrays

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Flow generated in deterministic lateral displacement (DLD) devices (Fig.1a) can be separated into controllable flow streams (or lanes) with a critical lane width, leading to a successful separation of rigid particles of different sizes [1]. In contrast to rigid particles, red blood cells (RBCs) are deformable, non-spherical in shape, and are subject to complex dynamics, including tank-treading, tumbling, and other states, which exhibit severe shape deformation and/or off-plane motion in flow [2]. These dynamical states affect the trajectory of RBCs in DLDs (Fig.1b) [3] and can be exploited for sorting cells with different intrinsic properties. Here, simulations are very promising for guiding and improving device design for a specific purpose.

In this study, we perform mesoscopic simulations of RBCs in DLD flow using the smoothed dissipative particle dynamics (SDPD) [4] method. We focus on the lateral displacement of RBCs in different dynamical states, which are sensitive to the flow rate and the viscosity ratio (C) between the suspending medium and a hemoglobin solution inside the cell.

Current results show a promising sensitivity of DLDs for RBC sorting under physiological viscosity conditions (C = 5). Thus, a particular row shift (Δλ) in DLD may result into zigzag mode for healthy RBC, while at the same time drive more rigid (e.g., diseased) states into displacement mode. Further insights for designing the post shape and post size will also be presented.

Fig. 1. (a) DLD device. (b) RBCs can flow above or under the separatrix. Adapted from [3]

References
Lateral migration of vesicle in bounded Poiseuille flow
a numerical investigation on effect of viscosity contrast
and initial position

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As the major component of blood, red blood cells contribute a large partition to hemorheology. For blood flow in capillaries, blood is often modeled as a mixture of Newtonian fluid (plasma) and vesicles encapsulating another fluid with higher viscosity (cytoplasm). The viscosity contrast of internal and external fluid can be affected by physiology and pathology condition, consequently it will change the effective viscosity of blood flow, which is an important indicator of blood. A 2-dim spectral boundary integral code is employed to investigate on how effective viscosity is perturbed by the variation of viscosity contrast. A diagram of single-vesicle lateral position versus initial position and viscosity is plotted. Non-linearity is observed and explained as a saddle bifurcation on critical initial position and viscosity contrast. Intrinsic viscosity of vesicle at lateral position is also investigated.
Cross-stream migration of asymmetric particles driven by oscillating shear

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We study the dynamics of asymmetric, deformable particles in oscillatory, linear shear flow. By simulating the motion of a dumbbell, a ring-polymer, and a capsule we show that cross-stream migration occurs for asymmetric elastic particles even in linear shear flow if the shear rate varies in time. The migration is generic as it does not depend on the particle dimension. Importantly, the migration velocity and migration direction are robust to variations of the initial particle orientation, making our proposed scheme suitable for sorting particles with asymmetric material properties. Further information is given also in [1].

References

Migration of soft microparticles in a modulated Poiseuille flow with the flow along the grooves

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We study the cross-streamline migration (CSM) of deformable particles in the limit of a vanishing Reynolds number in a spatially modulated planar Poiseuille flow with the flow along the grooves. By using Stokesian-dynamics simulation of 2D ring polymers, 3D capsules and red blood cells, we find besides a cross-streamline migration to the center between the two modulated confining plates, also CSM parallel to the confining plates to positions of minimal or maximal channel plate distances (positions of zero shear rate). The migration velocity depends for instance on the modulation amplitude as well as on the particle’s elasticity. The results obtained by Stokesian dynamics are confirmed by the Lattice-Boltzmann Method for arbitrary wall modulations. Our study suggests that the flow generated between wavy boundaries may be exploited for the separation of particles with different properties in microfluidic channels.
Migration reversal of soft particles in vertical flows

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Non-neutrally buoyant soft particles in vertical microflows are investigated. We find for light soft particles in downward Poiseuille flow cross-streamline migration (CSM) to off-center streamlines and for heavy particles CSM to the center. In both cases a reversal of the vertical flow direction and the related shear gradient causes a reversal of the migration direction. This gravitational driven CSM of soft particles occurs also in linear shear flows: heavy (light) particles migrate antiparallel (parallel) to the shear gradient. The surprising, flow-induced migration (reversal) is characterized by simulations and analytical approximations, confirming our plausible explanation of the effect. This might be applied for separating particles.
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