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# Large scale simulation of red blood cell aggregation in shear flows

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ABSTRACT

Aggregation of highly deformable red blood cells (RBCs) significantly affects the blood flow in the human circulatory system. To investigate the effect of deformation and aggregation of RBCs in blood flow, a mathematical model has been established by coupling the interaction between the fluid and the deformable solids. The model includes a three-dimensional finite volume method solver for incompressible viscous flows, the combined finite-discrete element method for computing the deformation of the RBCs, a JKR model–Johnson, Kendall and Roberts (1964–1971) (Johnson et al., 1971) to take account of the adhesion forces between different RBCs and an iterative direct-forcing immersed boundary method to couple the fluid–solid interactions. The flow of 49,512 RBCs at 45% concentration under the influence of aggregating forces was examined, improving the existing knowledge on simulating flow and structural characteristics of blood at a large scale: previous studies on the particular issue were restricted to simulating the flow of 13,000 aggregative ellipsoidal particles at a 10% concentration. The results are in excellent agreement with experimental studies. More specifically, both the experimental and the simulation results show uniform RBC distributions under high shear rates (60–100/s) whereas large aggregation structures were observed under a lower shear rate of 10/s. The statistical analysis of the simulation data also shows that the shear rate has significant influence on both the flow velocity profiles and the frequency distribution of the RBC orientation angles.



## 1. Introduction

The red blood cell (RBC, also referred to as erythrocyte) is the most common type of cell occurring in human blood and occupies approximately 45% of the total blood volume for man and 40% for women. RBCs in a healthy state have a biconcave shape with a diameter of 6–8 μm, a thickness of about 2 μm at the edges and about 1 μm at the centre (Baskurt, 2007). The RBC aggregation, which is the mechanism that greatly influences the non-Newtonian properties of blood (Baskurt et al., 2011), occurs when the shear forces are low and cells attract each other to form rouleaux (structures resembling coin piles), larger aggregates and networks of aggregates. RBC aggregation can cause complications in health related issues, therefore, the investigation of deformability and aggregation of RBCs in blood flow can be a promising tool for the better understanding and diagnosis of many diseases in clinical medicine.

Due to the complex mechanism of fluid–structure interaction, the theoretical analysis of RBCs or capsules are difficult and is usually limited to simple geometries and small deformations (Barthesbiesel, 1980) and an alternative approach–numerical simulation–has attracted much attention. Numerical methods suitable for RBC simulation include the boundary element method (BEM) (Pozrikidis, 1995, 2003) and the immersed boundary method (IBM) (Peskin and Mcqueen, 1980; Shi et al., 2012) together with a fluid solver based on either the finite volume method (FVM), the finite element method (FEM) (Liu et al., 2006) or the lattice Boltzmann method (LBM) (Feng and Michaelides, 2004; Sui et al., 2008; Zhang et al., 2008). In recent years, a family of particle-based methods has been used for such simulations, for example multi-particle collision dynamics (Gompper et al., 2009), smoothed particle hydrodynamics (Hosseini and Feng, 2009), and dissipative particle dynamics (Pivkin and Karniadakis, 2008; Pan et al., 2011; Quinn et al., 2011). As for the mechanism of aggregation of the RBCs, there are two co-existing models for the description of the mechanism: the bridging model and the depletion model (Chien and Jan, 1973; Neu and Meiselman, 2002). RBC aggregation has been incorporated into various fluid–structure interaction numerical models studying blood flow and Liu and Liu

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(2006) presented a three-dimensional model that couples the Navier–Stokes equations with cell interaction. The maximum number of RBCs involved in the simulation of the aforementioned study was 24. In yet another study by Zhao et al. (2012), the flow of 54 non-aggregating RBCs and 10 platelets was considered and the results showed that the migration of platelets towards the channel walls was affected by the local fluctuations of the medium fluid. Fedosov et al. (2011b) explicitly simulated the aggregation process of multiple RBCs successfully using dissipative particle dynamics (DPD) and predicted blood rheological properties.

To-date, most simulations have tended to target a relatively small number of cells (Kitano, 2002), possibly due to their complex nature. Such complex systems involve not only different categories of cells, but also very large numbers of simple and identical elements interacting to produce 'complex' behaviour. A relatively large simulation has been attempted by Chesnutt and Marshall (2009), who simulated the transport and aggregation of 13,000 RBCs using a discrete element method, during which the RBCs were modelled as rigid ellipsoidal particles and deformation was neglected. The intrinsic complexity of biological systems requires a closer combination between experimental and computational approaches (Doyle, 2001; Kitano, 2002). This again requires large scale simulations because experiments usually measure the macroscale effects with large quantities of cells (Doyle, 2001; Endy and Brent, 2001; Kaliviotis and Yianneskis, 2008; Dusting et al., 2009;). In this paper, computational research on RBC aggregations is presented in which the large scale nature of the computations have enabled direct comparisons of macro-scale aggregation features with experimental observations. To the knowledge of the authors, this is the first study set to examine numerically the flow and the changes in the structural characteristics of blood, caused by the phenomenon of RBC aggregation, at a large scale. In previous studies, where a very large number of RBCs have been used more than 100,000 RBCs in the study of Rahimian et al. (2010) no aggregative forces were taken into account. In addition, in the present study the simulated structural characteristics are directly compared to experimental cases.

## 2. Theory and methods

### 2.1. A solver for incompressible viscous flow

To simulate incompressible viscous flow, an in-house Computational Fluid Dynamics (CFD) code, called CgLes (Thomas and Williams, 1997) has been used. CgLes is a three-dimensional fluid solver with second order accuracy in both time and space and is based on a finite volume formulation. The capability of CgLes to simulate both laminar and turbulent flows has been extensively verified (Ji et al., 2012; Xu et al., 2012). The Navier–Stokes equations for incompressible viscous flows read:

$$\partial_t \mathbf{u} + \mathbf{u}\nabla\mathbf{u} = -\nabla p + \mu\nabla^2\mathbf{u} + \mathbf{f} \tag{1}$$

$$\nabla\mathbf{u} = 0 \tag{2}$$

where $\mathbf{u}$ is the vector of fluid velocities, $p$ is the pressure normalised with the fluid density and $\mathbf{f}$ is a body force term that can include the contribution from the immersed boundary method which will be described in the following sections. For time-stepping, a Crank–Nicolson scheme was used for the diffusion term and the convection term is discretised using an explicit two-step Adams–Bashforth scheme in order to retain the solver's capability for extension to higher Reynolds numbers.

### 2.2. Modelling the deformation of the RBCs

The combined finite-discrete element method (FEM–DEM) was used to simulate the movement and deformation of the RBCs under the various forces developed in the fluid (Munjiza and Wiley, 2004; Munjiza et al., 2011). The static shape of a healthy human RBC can usually be described with a biconcave discoid shape in which the $x, y$ coordinates of the cross-sectional profile of a RBC with a radius of $R=1$ follows the following function (Evans and Fung, 1972):

$$y = 0.5(1-x^2)^{(1/2)}(a_0 + a_1x^2 + a_2x^4), \quad -1 \le x \le 1 \tag{3}$$

where $a_0 = 0.207$, $a_1 = 2.002$, $a_2 = -1.122$.

In the present study, the RBC membrane was treated as a thin solid shell with a natural bending stiffness of $E_B = 1.82 \times 10^{-11}$ dyn cm. The membrane of a normal RBC is usually treated as a hyperelastic material which is usually modelled using either the neo-Hookean model (Huang et al., 2012) or the Mooney–Rivlin model (Liu and Liu 2006; Pozrikidis, 2005). The strain energy density functions of the neo-Hookean model $W_{nH}$ and the Mooney–Rivlin model $W_{MR}$ are:

$$W_{nH} = C_1(I_1-3); \quad W_{MR} = C_1(I_1-3) + C_2(I_2-3) \tag{4}$$

where $C_1$ and $C_2$ are empirically determined material constants, and $I_1$ and $I_2$ are the first and the second invariants of the unimodular component of the left Cauchy-Green deformation tensor. Note that both the models can be reduced to a Hookean model for a linear elastic material when the deformation is very small (Munjiza and Wiley, 2004). The Mooney–Rivlin model was adopted in this paper because of its greater extendibility to more complex materials. Please see the appendix for details.

### 2.3. Coupling the fluid and the soli interaction

To couple the fluid motion and solid deformation, an immersed boundary (IB) method (Peskin and Mcqueen, 1980) was used to link the interface between the fluid and the solid, both of which have independent meshes. By introducing a force term into the fluid's momentum equations, the immersed boundary points become non-slip boundary points. A spatially second order direct-forcing scheme (Ji et al., 2012) was used for the implementation of the immersed boundary method and a volume correction was introduced to maintain the conservation of RBC volume and surface area. Please see the appendix for details.

### 2.4. Modelling the interaction between RBCs

The adhesive force causing aggregation is short-ranged and originates from molecular forces, such as van der Waals attractions (Kendall and Stainton, 2001). The strength of the adhesion between two cells can usually be described by the adhesion work,$\sigma$, which is the work required to separate two adhered cells. The JKR model–Johnson, Kendall and Roberts (1964–1971) (Johnson et al., 1971) was used in the present study to compute the adhesion forces as this has proved to be applicable to the adhesion of living cells (Chu et al., 2005). The model provides the relationship between the adhesive force $F$ and the penetration depth $h$ via functions described in the following equation:

$$F = \frac{Ka^3}{R} - \sqrt{6\pi\sigma K a^3}, \quad h = \frac{a^2}{R} - \frac{2}{3}\sqrt{\frac{6\pi\sigma a}{K}} \tag{5}$$

where $R$ is the radius of the tip, $a$ is the contact area radius, $\sigma$is the work of adhesion and $K$ is the effective Young's modulus. For each element in contact, the adhesive force $F$ is computed and distributed to the nodes involved.

## 3. Validation and verification

### 3.1. Validation of the deformation of a single RBC

In order to verify the deformation of a RBC, the simulation of the stretching of a single RBC was carried out and numerical results were compared against experiment (Dao et al., 2003; Mills et al., 2004). Both the Mooney–Rivlin model and the neo-Hookean models were tested and compared. For the neo-Hookean model, a modulus $C = 2.57 \times 10^6$ dyn/cm$^2$ was used and for the Mooney–Rivlin model, moduli $C_1 = 2.57 \times 10^6$ dyn/cm$^2$ and $C_2 = 2.57 \times 10^5$ dyn/cm$^2$ were used following the highly related work of Skalak et al. (1973) and Liu and Liu (2006). The predictions by both the neo-Hookean model and the Mooney–Rivlin model agree closely with the published data (Dao et al., 2003; Mills et al., 2004) for both the axial and traverse deformations, (see Fig. 1).

### 3.2. Validation of the adhesive part of the model: disaggregation of two RBCs in a shear flow

The simulation results of the interaction between two individual RBCs were verified against the experiments of Chien et al. (1977), who investigated the shear stress needed to disaggregate a