Characterization of a dedicated mechanical model for red blood cells: numerical simulations of optical tweezers experiment

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1. Introduction

Red blood cells (RBCs) are biological cells devoid of nucleus. The complexity of these cells resides in their membrane which exhibits a microstructure consisting of a cytoskeleton resisting shear, and a quasi-incompressible lipid bilayer which provides a bending stiffness.

The characterization of a mechanical model of the RBCs membrane appears to be complicated but is of major interest. Indeed, numerical simulations of fluid-structure interactions of flowing RBCs have to rely on a realistic modeling of the membrane to provide accurate predictions of cells deformation. An effective way to characterize such a model is to compare numerical data coming from simulations with experimental data. Several experimental methods enable the investigation of the mechanical behavior of RBCs. Among these methods, the optical tweezers experiment provides a useful means for the analysis of the single cell mechanics under a variety of well-controlled stress-states.

In the present study, we aim at characterizing a mechanical model dedicated to RBCs. A number of existing models are investigated through optical tweezers simulations in order to determine the best suited one.

2. Methods

Optical tweezers simulations are performed using the YALES2BIO solver (http://www.math.univ-montp2.fr/~yales2bio/). This computational code embeds a finite-volume flow solver (Moureau et al., 2011) within which the front tracking immersed boundary method is implemented (FT-IBM), enabling numerical resolution of the fluid-structure interaction problem (Mendez et al., 2014).

The RBC membrane is supposed to be an infinitely thin hyperelastic surface which resists shear and area dilatation (Charrier et al., 1989). The internal and the external fluid are represented by a unique incompressible fluid of variable properties. Navier-Stokes equations are solved over a fixed Eulerian unstructured grid. The fluid velocity is interpolated from the fluid grid to the membrane vertices. In the FT-IBM framework, the membrane presence is taken into account by adding source terms to the Navier-Stokes equations, which mimic the force exerted by the membrane on the fluid.

The mechanical deformation imposed by the optical tweezers device is simulated by applying a force to two opposite regions corresponding to the contact area between the RBC and the 4.12µm diameter silica micro beads.

3. Results and Discussion

Optical tweezers simulations are performed using three hyperelastic models: the Neo-Hookean model, the Yeoh model, and the model dedicated to RBCs proposed by Skalak (Skalak et al., 1973).

The Neo-Hookean model is a one-parameter model in which shear and area dilatation resistances are taken into account through the shear modulus G_s (see Mills et al., 2004). The Yeoh model is a two-parameter model defined by the shear modulus G_s, and the non-linear modulus C_3 (see Mills et al., 2004). And the Skalak model enables to manage independently shear resistance and area dilatation resistance by means of the shear modulus G_s and the area dilatation modulus G_a, with $C = G_a / G_s$.

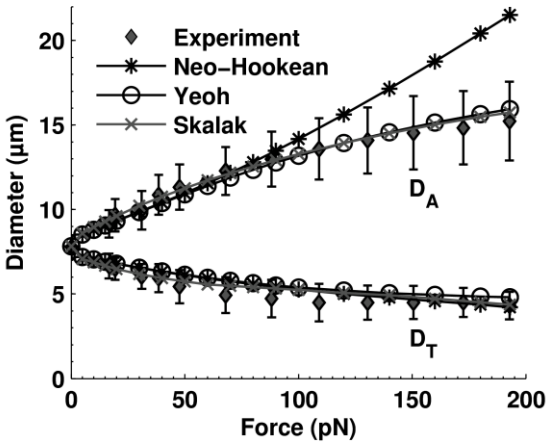

Figure 1 Axial (D_A) and transverse (D_T) diameters of RBCs stretched by optical tweezers. Comparison with experimental data (Mills et al., 2004).

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Figure 1 shows the mechanical response of a RBC stretched by optical tweezers, as a function of the applied force. The parameters used for Neo-Hookean and Yeoh models are the same as those used by Mills et al. (2004), i.e. $G_s = 7.3$ μN/m and $C_3 = G_s/30$. When using the Skalak model, the shear modulus is reduced by 50%, which leads to $G_s = 3.65$ μN/m and the constant C = 100. All tested models accurately capture experimental trends over the range of 0-88 pN, but the Neo-Hookean model deviates gradually after the 88 pN load, showing a strain-softening behavior under large deformation. Both the Yeoh model and the Skalak model provide accurate predictions of diameters over the entire range of experimental data. This indicates that they both transcribe well the strain-hardening behavior of RBCs under large deformation.

Area variations of the RBC membrane are reported in Table 1. A variation of 31% is obtained when the membrane is modeled by the Yeoh model. This observation is not consistent with the behavior of the RBCs for which a 3% to 4% increase in surface area results in cell lysis (Mohandas et al., 2008). The Skalak model enables to restrain the area variation, which is reasonably low for both C = 10 and C = 100. It is noted that the constant C strongly impacts the area variation observed during cell stretching, but does not influence the obtained cell diameters (not shown).

Model	Yeoh	Skalak (C=1)	Skalak (C=10)	Skalak (C=100)
ΔA (%)	30.9	12.7	2.1	0.3

Table 1 Area variation of the RBC membrane obtained for the maximal imposed force considering different mechanical models.

Figure 2 shows the equilibrium shape of the RBC using the Skalak model, for C = 1 and C = 100. The shapes differ according to the chosen constant C. Indeed, for C = 1, the RBC stretching induces the onset of a fold over the entire length of the RBC (which can be also observed with the Yeoh model). When the constant C is increased, area change restriction prevents the onset of the folding which gradually disappears.

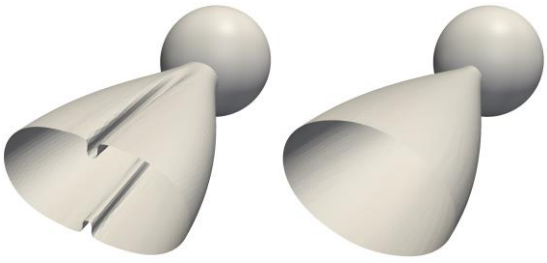

Figure 2 Half of the equilibrium shape of the RBC for the maximal imposed force: (left) Skalak model for C=1, (right) Skalak model for C=100.

4. Conclusions

Hyperelastic models designed to predict mechanical deformation of RBCs are investigated through optical tweezers simulations. A first analysis of the diameters of the stretched RBC allows to push aside the Neo-Hookean model, which predicts a strain-softening behavior under large deformation. In contrast, both the Yeoh model and the Skalak model reproduce well the strain-hardening behavior of RBCs. A second analysis of the membrane area variation shows the limitations of the Yeoh model, which enables non-physiological area increases.

The Skalak model appears to be the fittest model to predict the mechanical behavior of RBCs. Further investigations have to be conducted in order to give evidences on the choice of the constant C. Although the constant C does not impact the prediction of the diameters of the cell subjected to stretching, it is shown to strongly influence the membrane area variation and the obtained equilibrium shape. This underlines the necessity of a complete analysis of the cell diameters, membrane area variation and cell equilibrium shape to evaluate the ability of the model to predict mechanical deformation of RBCs.

Additional information provided by the experiment concerning the membrane area variation of the stretched RBC, and the folding which appears for low values of the constant C (close to C = 1), would be very helpful to calibrate the Skalak model.

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