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Analysis of laser beam scattering by an ensemble of particles modeling red blood cells in ektacytometer

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ABSTRACT

Using a simple theoretical model we have obtained approximate relations between the characteristics of particles modeling red blood cells and the parameters of the diffraction pattern produced by a laser beam diffracted in the ektacytometer. Laser beam diffraction by an ensemble of transparent elliptical discs with different shapes is considered. We have shown a feasibility of measuring certain parameters of the particles distribution in shapes by using the laser ektacytometry technique.

1. Introduction

An important microrheological parameter of red blood cell (RBC) is its deformability, defined as a measure of the cell ability to change its shape under action of an external force. This parameter essentially affects the RBCs motion through capillaries. An actual problem of bio-medical diagnostics is evaluation of RBCs deformability in various conditions.

Laser diffractometry of RBCs (ektacytometry) [1–3] is one of the techniques used to solve this problem. In an ektacytometer, an RBC suspension is placed in a gap between walls of two transparent coaxial cups, one of which is fixed while the other can be rotated at varying rates (so called Couette cell). Rotation of the cup causes the fluid flow and appearance of shear stress, which deforms the RBCs. To make the particles deformation visible, the erythrocytes suspension is illuminated with a laser beam. As a result, a diffraction pattern arises on the observation screen. This pattern provides information about shapes of the particles under investigation.

As a rule, the diffraction pattern contains a bright central maximum and a number of weakly distinguished interference lines, either circular or elliptic in shape depending on the extent of shear-induced deformation of RBCs. This testifies to the fact that erythrocytes in shear flow acquire ellipsoidal shape. The diffraction pattern is recorded by a video camera and then is transmitted to a computer. The computer selects the points at the observation screen where the scattered light intensity has a certain fixed value. Usually, in conventional ektacytometers these points are located at the border of the central diffraction maximum [4]. A line obtained in this way is referred to as an isointensity curve. Its shape is approximated by an ellipse. The ratio of the ellipse axes as

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a function of the shear velocity or shear stress in the Couette cell is used as a quantitative measure of the deformability of RBCs in the tested suspension.

An important question is how the characteristics of RBCs are related to the parameters of the diffraction pattern obtained in the ektacytometer. Answering this question requires a theoretical model, which would enable one to calculate the laser light diffraction by a single erythrocyte as well as by an ensemble of erythrocytes. At present, there are several numerical methods that can be used to solve this problem with high accuracy. These are discrete-dipole approximation (DDA) [5,6], finite-difference time-domain method (FDTD), T-matrix method [7], anomalous diffraction approximation [8,9], ray-wave approximation [10,11], and others. However, the results of numerical calculations are very concrete and that makes it difficult to find functional relations between the input and output parameters of the problem.

On the other hand, for the experimental data analysis, approximate analytical relations, connecting the characteristics of erythrocytes with the parameters of the diffraction pattern, may be very useful. An example of such relation is a formula, found in [12], that connects the erythrocytes size dispersion parameter δ^2 with the visibility of the diffraction pattern v: $v = 1 - 76\delta^2$. This relation is obtained with a model, which considers an erythrocyte as a circular transparent disc, and calculates the laser beam scattering by using the diffraction integral [13,14]. In this work, the scatter of the particles in sizes was assumed to be relatively small ($\delta^2 \ll 1$).

Generally speaking, in a population of RBCs different cells possess different abilities to deform. This is a reason to consider the deformability as a statistical characteristic feature of an ensemble of RBCs and to describe it by a distribution function, mean value and dispersion. In this paper, we consider the problem of how a scatter of particles in shape affects the diffraction pattern observed and processed in the ektacytometer. We believe that answering this question would enable one to evaluate the possibilities of laser ektacytometry as a tool for measuring the parameters of RBCs scatter in the deformability.

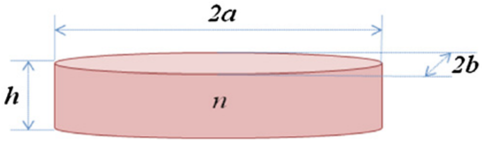

Fig. 1. Elliptical disc modeling an erythrocyte deformed in a shear flow. Parameters of the model: n—relative refractive index, (a, b, h)—geometrical sizes.

2. Effect of particles distribution in shapes on the diffraction pattern

Experimental data of laser ektacytometry, as well as images obtained with a microscope [15,16], show that normal erythrocytes in a shear flow acquire a shape close to an ellipsoid. To make analytical estimates of angular distributions of the intensities of light diffracted by such particles, we will model the erythrocytes by plane transparent discs with bases of elliptical shape (Fig. 1).

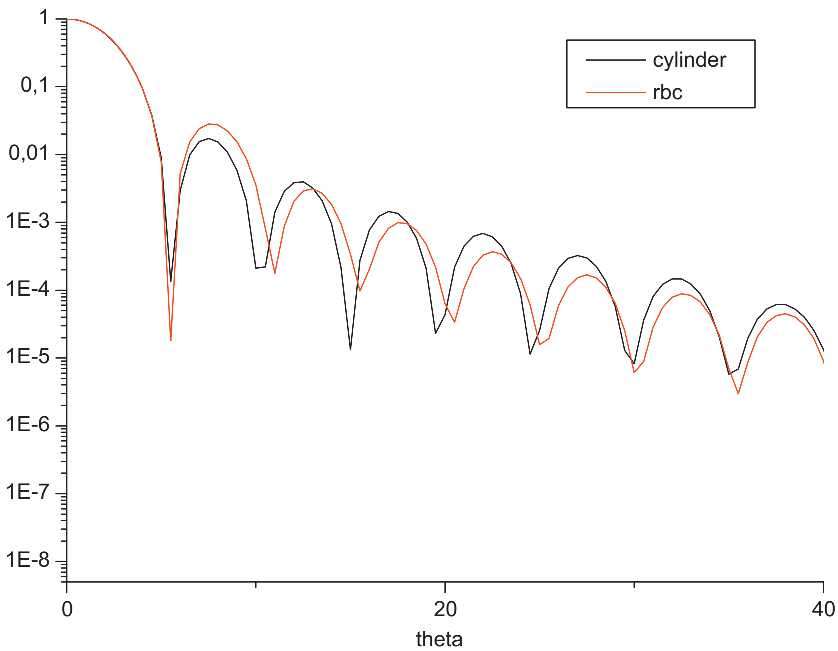

Fig. 2. Angular distributions of scattered light intensity for the cases of laser beam scattering by a cylinder (plane disc) and biconcave disc modeling a red blood cell. The distributions are calculated using the ADDA code [5]. The incident laser beam is supposed to be parallel to the particles symmetry axes. The particle diameter is 8 μm, laser wavelength – 0.633 μm, and relative refractive index – 1.05. The cylinder height – 1.5 μm, minimum thickness of the biconcave disc – 1 μm, and maximum thickness – 2 μm (achieved at a distance of about 2.8 μm from the disc center). The scattered light intensity is normalized to the intensity of the central diffraction maximum.