Influence of the Endothelial Surface Layer on the Wall-induced Migration of Red Blood Cells

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Abstract

The endothelial lining of blood vessels presents a large surface area for exchanging materials between blood and tissues. The endothelial surface layer (ESL) plays a critical role in regulating vascular permeability, hindering leukocyte adhesion as well as inhibiting coagulation during inflammation. Changes in the ESL structure are believed to cause vascular hyperpermeability and induce thrombus formation during sepsis. In addition, ESL topography is relevant for the interactions between red blood cells (RBCs) and the vessel wall, including the wall-induced migration of RBCs and formation of a cell-free layer. To investigate the influence of the ESL on the motion of RBCs, we construct two models to represent the ESL using the immersed boundary method in two dimensions. In particular, we use simulations to study how lift force and drag force change over time when a RBC is placed close to the ESL as the thickness, spatial variation, and permeability of the ESL vary. We find that spatial variation has a significant effect on the wall-induced migration of the RBC when the ESL is highly permeable and that the wall-induced migration can be significantly inhibited by the presence of a thick ESL.

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Significance: The interplay between the endothelial surface layer (ESL) and blood plays a pivotal role in maintaining a healthy vascular environment. However, it is not yet well-understood how the disturbances in ESL structure affect the microcirculation. In this work, we show that the wall-induced migration of red blood cells (RBCs) may be significantly inhibited by changes in the thickness and spatial variation of the ESL. By combining our model with medical images showing the changes in the ESL that occur during sepsis, we demonstrate how the ESL may affect the formation of the cell-free layer (CFL) in disease states. The work highlights significant potential influence of the ESL on microcirculation, wall-induced migration of RBCs and the formation of the CFL.
I. INTRODUCTION

Blood flow is a key determinant in numerous pathologies such as tumor vascularization [1], sickle cell anemia [2], and atherosclerosis [3]. Along with plasma, red blood cells (RBCs) are major components of blood, occupying 40% – 45% of its volume [4]. At rest, a healthy RBC has a biconcave shape with a diameter of 8 $\mu$m and a width of 2 $\mu$m and is highly deformable, which allows them to pass through capillary vessels with a diameter as small as 2.7 $\mu$m [5]. Due to the high occupancy of RBCs, the blood flow is, therefore, greatly influenced by the dynamics of RBCs.

Fluid-mediated interactions between circulating RBCs and vessel walls lead to a non-uniform distribution of cells and ESL-dependent resistance to flow. *In vitro* experiments of blood flow in narrow glass tubes reveal the cross-stream migration of RBCs, leading to the formation of a cell-depleted layer or cell-free layer (CFL) near the tube wall [6–8]. Upon formation of the CFL, RBCs acquire a lift force and migrate away from the tube wall. Such a phenomena is often termed wall-induced migration. *In vivo* experiments of blood perfused through microvessels confirm the formation of the CFL next to the vessel wall [9–11]. The spatial and temporal variation of the CFL is shown to be nonaxisymmetrical and positively related to the vessel diameter [9]. Notably, the blood flow resistance measured *in vivo* is higher than that measured *in vitro*. The presence of the endothelial surface layer (ESL) in microvessels is believed to be a contributing factor to the substantial increase in flow resistance and potentially affect the formation of the CFL. However, the ESL is often omitted in studies of the wall-induced migration of RBCs [12, 13].

The endothelial surface layer is made of two semi-distinct layers of membrane-bound macromolecules. The inner part comprises a thin layer (0.05 – 0.4 $\mu$m) of molecules dominated by glycoproteins and proteoglycans that are anchored to the endothelial cells. This layer has been observed to have a quasi-periodic matrix structure [14] with brush-like configurations of fibers [15, 16]. The outer layer consists of a complex array of soluble plasma proteins and glycosaminoglycans that extends approximately 0.5 $\mu$m from the inner layer [17]. Previous studies suggest that the whole layer, in general, resists compression and flattening upon interaction with RBCs through analyzing the variation of the osmotic pressure [18, 19]. The ESL has been shown to impede the flow of plasma through modeling the ESL as a porous medium using Brinkman approximation and examining its hydraulic resistivity [18, 20].
recent study that models the ESL-RBC interaction using dissipative particle dynamics revealed the ESL’s additional functionality as a mechanotransducer [21].

In this work, we followed the approach used in previous studies [22, 23] develop a two-dimensional computational model to study the effect of the ESL on the wall-induced migration of RBCs and in particular the formation of the CFL during sepsis. To understand the role of the ESL structure in the motion of the RBC near the layer under pathological conditions, we developed two models that capture the ESL at different levels of resolution. In the first model, the topography of the ESL is coarse-grained into a smooth wave (Fig. 1A), whereas in the second model, the brush-like structure of the ESL is represented explicitly (Fig. 1B). An open source code repository containing MATLAB code used for the simulations may be found at https://github.com/phzhang0616/RBC-ESL_IB_Simulation.git.

We use our models to study computationally how the drag and lift force experienced by a RBC vary as the spatial variation of the layer is changed. Using the detailed model, in which the structure of the ESL is represented explicitly, we find that the wall-induced migration of the RBC is hindered as the density of bundles increases for highly permeable ESLs. In contrast, for impermeable ESLs we find that the wall-induced migration of the RBC remains largely unchanged. We further show that the RBC is less likely to migrate away from the layer as the thickness is increased, and the permeability of the ESL does not appear to have a strong influence on the wall-induced migration as the thickness varies. We reproduce these results using a coarse-grained model, in which the ESL is described as a sinusoidal wave. We then extract ESLs from medical images provided in [24] and apply the
coarse-grained model to study the formation of the CFL in simulations including only RBCs or both RBCs and leukocytes. In both cases, we find that the thickness of the resulting CFL increases significantly in the disrupted scenario, leading to an inverse relation between the CFL thickness and the vessel diameter. The nonaxisymmetric nature of the CFL is preserved in the healthy and disrupted cases in the system containing RBCs and leukocytes. In the images analyzed here we find the CFL on the top side to be consistently thicker than the bottom CFL. Our work highlights the importance of the ESL in studying microcirculation, and suggests a possible unexpected role for the ESL in controlling the formation of the CFL in different pathologies.

II. MATERIAL AND METHODS

Mathematical Model of Blood Flow

We let $\Omega \subset \mathbb{R}^2$ be a bounded domain representing the blood vessel filled with blood, which is incompressible, Newtonian, and contains RBCs and ESLs. In all the simulations, we assume $\Omega$ is a rectangle. The dynamics of blood flow are described via the unsteady Stokes equation

$$\text{Re} \frac{\partial \mathbf{u}}{\partial t} + \nabla p = \Delta \mathbf{u} + \mathbf{f}$$

$$\nabla \cdot \mathbf{u} = 0,$$

equipped with boundary conditions as follows

$$\mathbf{u} = \mathbf{0} \text{ on the top and bottom of } \Omega,$$

$$\mathbf{u} \text{ periodic in the } x \text{ direction.}$$

In Eq. II.1 $\mathbf{u}(x, t)$ denotes the fluid velocity, $p(x, t)$ is the pressure, both defined in terms of the Eulerian coordinates, $x = (x, y)$, and Re is the Reynolds number. The force, $\mathbf{f}(x, t)$, applied to the blood is given by

$$\mathbf{f}(x, t) = \mathbf{f}_{\text{IB}}(x, t) + \mathbf{f}_{\text{Body}}(x, t),$$

where $\mathbf{f}_{\text{IB}}(x, t)$ is the force induced by the presence of RBCs and ESLs (see Methods) and $\mathbf{f}_{\text{Body}}(x, t)$ is a body force applied to the blood to establish the flow given by

$$\mathbf{f}_{\text{Body}}(x, t) = (\sin(\pi y), 0)$$

as used in [25]. In the absence of the immersed objects, this establishes a unidirectional flow in the domain.
Elasticity Model for the RBC Membrane

We use a two-dimensional elastic spring model to describe the elasticity of the immersed RBCs, which is represented on a moving Lagrangian grid with coordinates \( q \in [0, L_R] \) and the corresponding Eulerian coordinates at time \( t \) are given by \( X(q, t) \). We assume that the membrane Lagrangian points are connected by springs having total elastic energy for stretching/compressing

\[
E_{\text{spring}} = \frac{k_s}{2} \int_0^{L_R} \left( \left\| \frac{\partial X(q, t)}{\partial q} \right\| - 1 \right)^2 dq, \tag{II.5}
\]

and by torsional springs with total energy for bending

\[
E_{\text{bend}} = \frac{k_b}{2} \int_0^{L_R} \left\| \frac{\partial^2 X(q, t)}{\partial q^2} \right\|^2 dq, \tag{II.6}
\]

The coefficients \( k_s \) and \( k_b \) are elastic stretching and bending constants respectively. When the RBC deforms, its surface area and volume remain relatively constant. To conserve the area of RBC in the model, we prescribe a penalty energy

\[
E_{\text{area}} = \frac{k_a}{2} (A(t) - A_0)^2, \tag{II.7}
\]

where \( k_a \) is an area-preserving constant, \( A(t) \) is the area of the RBC at time \( t \) and \( A_0 \) is the target area. In all simulations \( k_a \) is picked so that the maximum loss of area of the RBC is less than 2%. The total Lagrangian force for the RBC membrane maybe be calculated via

\[
F_{\text{RBC}} = - \left[ \frac{\partial E_{\text{spring}}}{\partial X} + \frac{\partial E_{\text{bend}}}{\partial X} + \frac{\partial E_{\text{area}}}{\partial X} \right], \tag{II.8}
\]

where \( \frac{\partial}{\partial X} \) is the Fréchet derivative.

Elasticity Model for the ESL

A key consideration of the model is the ESL geometry. Due to the lack of direct measurements of the mechanical properties of the ESL, we follow the information provided in previous studies \([18, 26]\) to model the ESL as an immobile structure. Observations of the restoration of ESL thickness within approximately one second following its compression by a passing white blood cell indicate that the layer is deformable but has a finite resistance to compression \([27]\). Following this evidence, we model the layer as an elastic structure. To study the influence of the ESL’s spatial variation and thickness on the near-wall motion of a RBC, we represent its structure using curvilinear boundaries on a Lagrangian grid with coordinates \( \hat{q} \in [0, N_E] \). We assume that there are two ESLs on the top
and bottom of the computational domain with the corresponding Eulerian coordinates given by \( \hat{X}_0(\hat{q}, t) \) and \( \hat{X}_h(\hat{q}, t) \) respectively. As the layer is mostly immobile, we use tether points to connect \( \hat{X}_0 \) and \( \hat{X}_h \) to virtual lines \( Z_0(t) \) and \( Z_h(t) \) by stiff springs with spring constants, \( k_{tether} \). The corresponding Lagrangian forces are

\[
F_{\text{ESL}}^0 = -k_{tether} \left( \hat{X}_0(\hat{q}, t) - Z_0(t) \right),
\]

\[
F_{\text{ESL}}^h = -k_{tether} \left( \hat{X}_h(\hat{q}, t) - Z_h(t) \right).
\]

To study the first model, in which a detailed representation of the ESL is used, we described a single ESL as bundles of fibers (see Figs. 1B and 4A). The corresponding elastic energy is defined identically as Eq. II.5. The root of each fiber is tethered to the vessel wall using tether points. In the simplified model, we coarse-grained the ESL into a continuous sinusoidal wave, which can be interpreted as an approximation to the variation of the ESL in the first model (see Figs. 1A and 6A). In this case, all the Lagrangian points representing the ESL are connected to tether points.

*Immersed Boundary Method*

To couple the models for blood motion, the RBC, and the ESL, we employ an immersed boundary method as in [28, 29]. We make use of the discretized Dirac delta functions to transfer information between the Eulerian and Lagrangian coordinates. The Eulerian force density in Eq. II.3 induced by the RBC and ESL is determined by the Lagrangian force density via

\[
f_{\text{IB}}(x, t) = \int_q F_{\text{IB}}(q, t) \delta(x - X(q, t)) \, dq,
\]

where \( F_{\text{IB}} = F_{\text{RBC}} + F_{\text{ESL}}^0 + F_{\text{ESL}}^h \).

The impermeable immersed object moves at a velocity \( U(q, t) \) equal to the local fluid velocity. This may be expressed in terms of the Dirac delta function by

\[
U_{\text{RBC}}(q, t) = \int_{\Omega} u(x, t) \delta(x - X(q, t)) \, dx,
\]

\[
U_{\text{ESL}}^{0,h}(\hat{q}, t) = \int_{\Omega} u(x, t) \delta \left(x - \hat{X}_{0,h}(\hat{q}, t)\right) \, dx.
\]

To study the effect of ESL’s permeability in the Results section, Eq. II.13 must be adjusted to account for porosity

\[
U_{\text{ESL}}^{0,h}(\hat{q}, t) = -\nabla \cdot n_{0,h} + \int_{\Omega} u(x, t) \delta \left(x - \hat{X}_{0,h}(\hat{q}, t)\right) \, dx,
\]
where \( \hat{n} \) is a unit normal vector to the ESL and \( U_{p}^{0,h} \) are defined as

\[
U_{p}^{0} = -k_{p}^{0} \frac{F_{ESL}^{0} \cdot \hat{n}_{0}}{\|\hat{X}_{0}\|}, \quad (\text{II.15})
\]

\[
U_{p}^{h} = -k_{p}^{h} \frac{F_{ESL}^{h} \cdot \hat{n}_{h}}{\|\hat{X}_{h}\|}. \quad (\text{II.16})
\]

Together, \( U_{p}^{0} \hat{n}_{0} \) and \( U_{p}^{h} \hat{n}_{h} \) correspond to the slip velocities given by Darcy’s law for the top and bottom ESL respectively [30–32]. Here we assume the porosity constants, \( k_{p}^{0} \) and \( k_{p}^{h} \), for the top and bottom layer are equal. As stated in [30, 31] the porosity constant \( k_{p} \) is derived from Darcy’s law [33]. In particular, \( k_{p} \) is proportional to the number density of the pores per unit length of the layer. That is, for a fixed layer thickness increasing the permeability is equivalent to increasing the number of pores, thus increasing the blood flux through the layer.

The model is spatially discretized as in [34] and temporally using a second-order accurate predictor-corrector time-stepping scheme as in [25]. In SI Section VII S1, we provide a detailed description of how the spatial discretization is formulated. As a validation of our model, in Results we simulate the wall-induced migration of a single deformable membrane and compare to prior results from [20].

To study the influence of the ESL structure on the motion of RBCs near the ESL, we performed simulations in dimensionless units on rectangular domain of size \([0, 2] \times [0, 1]\) unless otherwise stated. The domain is partitioned into a Cartesian mesh of \(256 \times 128\) grid points. As shown in SI VII S2, the simulation results are sufficiently well-resolved with this choice of mesh size. The Lagrangian point spacing is picked to be half of the domain mesh spacing. The corresponding discretized energies of Eqs. II.5-II.7 used in simulations are given in SI Section VII S13. Simulations were run till \( T = 50 \) so that the center of mass of the RBC reaches a steady motion in the vertical direction for the impermeable ESL (see Fig. 4A and Fig. 6A). The lift and drag were calculated as functions of time by summing the forces at each immersed boundary point at a fixed saving time step and taking the opposite sign of that value as described in [35]. With this approach the lift force is defined as the force acting perpendicular to the direction of flow, and the drag is defined as the force pushing the RBC moving along the flow direction horizontally. To minimize the effect of the ESL topography on the drag and lift force, we calculated both forces by averaging over four initial conditions that either uniformly sample the distance between two neighboring bundles for the detailed model or one wavelength of the sine wave for the coarse-grained model.
III. RESULTS

Validity of unsteady Stokes for blood flow and the membrane elasticity model

As a benchmark, we first applied the coarse-grained model (described in further detail later on in Results) with dimensional parameters to the problem setup described in [20] focusing on the effects of porous wall layer on the motion of a single deformable membrane near microvessel walls. We examine two scenarios corresponding to impermeable ESL layers and permeable layers with a hydraulic resistivity of $\kappa = 10^{11}\text{N}\cdot\text{s}/\text{m}^4$ [20]. For impermeable layers, the computational domain is a rectangular channel of 16µm in length and 8µm in width equipped with a periodic boundary condition in the horizontal direction and a no-slip boundary condition in the vertical direction. In this case, ESLs are modeled explicitly as solid walls and tether points are not used. For permeable layers, the computational domain is a rectangle of length 20µm and width 10µm with the same boundary conditions as in the impermeable case. An ESL of width 1 µm is assumed and is modeled as a straight line as in [18, 20]. In both cases the initial configuration of the membrane is a circle of radius 2.66 µm and is placed with its center 0.9 µm from the center-line of the channel. A body force of the form described above in Eq. II.4 is applied and the magnitude is chosen so that the maximum flow velocity matches the one in [20] (approximately 0.8mm/s) in the absence of the membrane with impermeable ESLs.

One fundamental difference between our approach and the one used in [20] is the model of permeability of the layer. In [20] the porous ESL is modeled using Brinkman’s approximation and the variation in the layer’s permeability is characterized by the hydraulic resistivity ($\kappa$). To determine the corresponding porosity constant, $k_p$, in our model, we estimated $k_p$ via $\kappa = \mu/k_p$ [31, 36]. Here $\mu$ is the fluid viscosity and is set to $10^{-3}\text{Pa}\cdot\text{s}$ as in [20]. We summarized in Table. S1 the parameters used in our simulations.

We compute the motion and deformation and the distance of the center of mass of the membrane from the center-line of the channel and compare them to those reported in Figure 2 and Figure 3 of [20]. We show in Figs 2 and 3 that the results of our coarse-grained model are initially in good agreement with [20], as the membrane migrates away from the top layer and moves towards the center-line of the channel. After 200ms however, the membrane continuously moves towards the center-line and reaches a steady state in our model whereas in [20] the center of mass of
the membrane appears to oscillate slightly (see Fig. 3). In comparing the deformation of the membrane between the impermeable and permeable case, we find that, similar to [20], the shape of the membrane remains largely unchanged using our coarse-grained model (see Fig. 2). We note that the deformation of the membrane using our two-dimensional model differs somewhat from that reported in [20], in that the parachute-like concavity on the trailing end of the membrane is less prominent in our simulations than in [20]. However, note that the resulting shapes are not expected to closely resemble those of a flowing RBC since the membrane is initialized as a circle in this simulation, whereas the equilibrium configuration of an RBC is known to be a biconcave disk, which has a significantly smaller reduced volume. While the circular reference configuration
is satisfactory for benchmarking our simulation setup in the context of a deformable membrane, when simulating RBC’s in the following sections we will use a biconcave disk initial configuration.

![Graph](image)

**FIG. 3.** Distance of center of mass of the RBC from the center-line of the channel. Results extracted from [20] using [37] are plotted in solid (impermeable) and dashed (permeable, $\kappa = 10^{11}\text{N}\cdot\text{s}/\text{m}^4$) lines. Results of using our coarse-grained model are plotted in circles (impermeable) and stars (permeable, $k_p = 10^{-2}\mu\text{m}^2$). Parameters are summarized in Table S1.

**Spatial variation of ESL has a minimal effect on the motion of RBC for impermeable layers**

Interactions between circulating RBCs and ESLs are central to immune [41] and inflammatory responses [42]. Following acute injury and inflammatory conditions such as sepsis, disruption of the ESL can be characterized by changes in ESL’s spatial variation and thickness [24]. To investigate the influence of ESL topography on the wall-induced migration of a RBC when it is within a close proximity to the layer, we use an immersed boundary method (see Methods). The model includes two ESLs with one on the top and one at the bottom of the channel and a single RBC. The biconcave shape of the RBC is generated using the parametrization given in [39]. We explicitly include the effect of layer’s spatial variation and thickness by modeling each ESL as a collection of elastic fiber bundles (Fig. 4A). We assume that there are 10 bundles evenly spaced across the domain. In our model of a healthy ESL, we include at most 5 fibers tethered to the root of the
| Parameter | Description                  | Value                     |
|-----------|------------------------------|---------------------------|
| Re        | Reynolds number              | 0.01 [25, 38]             |
| $k_s^{\text{RBC}}$ | Elastic spring constant | 3 μN/m [39]               |
| $k_b$     | Bending constant             | $2 \times 10^{-19}$ N·m [23, 40] |
| $k_a$     | Area preserving constant     | 185 N/μm²                 |
| $k_{\text{tether}}$ | Tether force constant | 3200 N/μm                |
| $k_p$     | Porosity constant            | 0, 0.01μm² [20]           |
| $\Delta x$, $\Delta y$ | Domain mesh spacing | 0.0078μm                  |
| $\Delta q$ | Lagrangian mesh spacing     | 0.0039μm                  |
| $\Delta t$ | Time-step size              | 0.0001s                   |

bundle consistent to the reported ESL structure [16]. The layer thickness, $h$, is defined to be the maximum height of a bundle. Spatial variation of the layer is given by the density of bundles and is defined to be the inverse of the distance between the roots of two neighboring bundles. In exploring the effect of density of bundles, we decrease the number of bundles until there are two bundles left, corresponding to an ESL depletion which occurs in sepsis and diabetes [17, 24, 26, 43].

We focus on the combined effects of ESL’s spatial variation, thickness and permeability on the migration profile of the RBC and as such, we have introduced two simplifications. First, the RBC is initialized with a 30 degree orientation (see Fig. S4A). Second, we do not use a large elastic spring constant to model stiff fibers but instead introduce tether forces. These simplifications decrease the computational complexity of the model and are not expected to significantly alter the conclusions. In particular, using a 30 degree orientation allows us to pick a relatively small simulation termination
FIG. 4. The effect of density of bundles in changing the drag and lift forces is more apparent for permeable ESL. (A) The position of the RBC is shown at indicated times as it moves through the channel (top) with the motion of the cell on top of the steam-line at \( T = 0.1 \) (bottom). (B and D) Time and initial condition averaged drag and lift force versus density of bundles for different values of permeability. (C and E) Time and initial condition averaged fraction of drag \( \frac{\langle \text{Drag} \rangle}{\langle \text{Drag} \rangle + \langle \text{Lift} \rangle} \) and lift force \( \frac{\langle \text{Lift} \rangle}{\langle \text{Drag} \rangle + \langle \text{Lift} \rangle} \) versus density of bundles. In all simulations, the thickness is fixed to be \( h = 0.1827 \). In panel B and D a 95% confidence intervals are plotted at each data point. Parameters are summarized in Table II.
time by minimizing the initial transition period, during which the RBC deforms and reorients before moving away from the wall. Moreover, explicitly modeling the stiffness of fibers through tether forces is a common approach to approximate solid objects in immersed boundary methods [44] that allows for a larger simulation time-step, and is not expected to significantly alter the results. In all simulations reported here, the RBC is initialized at least four grid points away from the layer. As such, the RBC is sufficiently far from the layer to resolve the flow using the standard immersed boundary method without lubrication corrections.

TABLE II. Parameters for the detailed model (Figs. 4 and 5)

| Parameter | Description                  | Value                |
|-----------|------------------------------|----------------------|
| Re        | Reynolds number              | 0.01 [25, 38]        |
| $k_s^{RBC}$ | Elastic spring constant   | 3 $\mu$N/m [39]     |
| $k_b$     | Bending constant            | $2 \times 10^{-19}$ N.m [39] |
| $k_a$     | Area preserving constant    | 185 N/$\mu$m²       |
| $k_{tether}$ | Tether force constant      | 3200 N/$\mu$m       |
| $k_s^{ESL}$ | Elastic spring constant  | 0.015 $\mu$N/m       |
| $k_p$     | Porosity constant           | 0 – 0.01 $\mu$m² [20] |
| $h$       | Thickness of one ESL        | 0.8464 – 1.7696 $\mu$m [26] |

As a first output of the model, we calculated the drag and lift force of the RBC averaged over time and four initial conditions that uniformly sample the distance between two neighboring bundles (Fig. S4A) as the density of bundles was increased. The absolute value of both the drag and lift are reported. Interestingly, when using different densities of bundles, we find that decreasing density of bundles has a minimal effect in the drag force, with a maximum increase of 1%, and the lift force, with a maximum decrease of 14%, when the layer is impermeable or less permeable,
corresponding to a healthy scenario. As the layer’s permeability is increased, corresponding to the damaged/disturbed ESL, decrease in the density of bundles leads to a more apparent decrease in the lift force, a maximum decrease of 25%, but the change in the drag force remains largely unchanged, a maximum increase of 1% (Fig. 4, B and D).

We quantified the wall-induced migration of the RBC by calculating the drag and lift over the total amount of force. Plots of such fractions over the density of bundles show that the ability of RBC of moving away from a highly permeable ESL is significantly inhibited as the density of bundles increases due to a decrease in the drag (Fig. 4C) and an increase in the lift (Fig. 4E) whereas the motion is less affected as the density of an impermeable ESL increases (Fig. 4, C and E). Taken together, we find that the spatial variation of ESL plays a more dominant role in affecting the migration of the RBC when the layer is highly permeable, but minimal when the layer is less permeable.

_A thicker ESL can slow down the migration of the RBC_

Our detailed ESL model makes it possible to capture physical features of the ESL similar to those observed in experiments. In addition to the spatial variation, another way in which the ESL becomes disturbed in pathological states is through the change in thickness. For instance, ESL degradation in diabetes leads to a thinner layer [24, 45] while a thicker ESL is observed during edema formation [43]. To determine the importance of the ESL thickness, we assume the density of bundles remains unchanged with the distance between the roots of two neighboring bundles fixed to be 0.2.

We calculated the time and initial condition averaged drag and lift and analyzed their magnitude. As expected, the ESL’s thickness is positively related to the drag and negatively related to the lift. For impermeable layers, increasing the ESL’s thickness increased the drag force by 32% and decreased the lift force by 15%. In this case, we find that the RBC is more prone to stay near the ESL due to an increasing amount of confinement. Unexpectedly, we find a similar change in the drag and lift force when the ESL is highly permeable. The drag force has a maximum increase of 29% and the lift force has a maximum decrease of 14% (Fig. 5, A and C). As before, we report these results by quantifying the wall-induced migration of the RBC. We find that for all physically-relevant choices of the layer permeability the drag force continuously increases as the thickness is
FIG. 5. The effect of thickness in changing the drag and lift forces is apparent regardless of the permeability. (A and C) Time and initial condition averaged drag and lift force versus layer thickness for different values of permeability. (B and D) Time and initial condition averaged fraction of drag ($|\langle \text{Drag} \rangle|/(|\langle \text{Drag} \rangle| + |\langle \text{Lift} \rangle|)$) and lift force ($|\langle \text{Lift} \rangle|/(|\langle \text{Drag} \rangle| + |\langle \text{Lift} \rangle|)$) versus thickness over all values of permeability. In all simulations, we assume the ESL is in a healthy condition corresponding to a density of bundles of 5. In panel A and C a 95% confidence intervals are plotted at each data point. Parameters are summarized in Table II.

increased and consistently dominates the lift force, causing the RBC to remain near the layer due to a lack of upward lift (Fig. 5, B and D).

*The coarse-grained model is sufficient to capture the important features of the ESL*

The preceding detailed model demonstrates a clear relation in how the drag and lift of a RBC depend on the ESL’s thickness as permeability varies. It also suggests that the effect of the ESL’s spatial variation is minimal when the ESL is impermeable. To develop a more computational
TABLE III. Parameters for the coarse-grained model (Figs. 6 and 7)

| Parameter | Description                    | Value                  |
|-----------|--------------------------------|------------------------|
| Re        | Reynolds number                | 0.01 [25, 38]          |
| $k_{s\text{RBC}}$ | Elastic spring constant                | 3 $\mu$N/m [39]         |
| $k_b$     | Bending constant               | $2 \times 10^{-19}$ N·m [39] |
| $k_a$     | Area preserving constant       | 185 N/$\mu$m$^2$       |
| $k_{\text{tether}}$ | Tether force constant                | 3200 N/$\mu$m         |
| $k_{s\text{ESL}}$ | Elastic spring constant                | 0.015 $\mu$N/m        |
| $k_p$     | Porosity constant              | 0 – 0.01 $\mu$m$^2$ [20] |
| $(A + h)$ | Thickness of one ESL           | 0.8464 – 1.7696 $\mu$m [26] |
| $a/(2\pi)$ | Spatial frequency              | 1 – 10                 |

efficient model that captures these important physical features of the ESL, we exploited the fact that
the RBC only interacts with the surface/outer region of the ESL to coarse-grain its structure into
a continuous sine wave of the form $y(x) = A\sin(ax) + h$ where $(A + h)$ represents the thickness of a
single ESL either on the top or at the bottom and the spatial variation of the ESL is characterized
by the spatial frequency, which is given by the frequency $a$ of the sine wave. Throughout this
work we fix the sinusoidal amplitude to be $A = 0.336\mu$m. To demonstrate the difference in the
computational cost between the coarse-grained and the detailed model, we simulated a dense case
of the ESL using both models with thickness fixed for $k_p = 0, 0.005,$ and $0.01\mu$m$^2$. As shown in SI
Section VII S3, the coarse-grained model is approximately twice as fast as the detailed model and
the speed-up factor increases for larger values of $k_p$.

As before, we calculated the drag and lift averaged over the course of the simulation for four
FIG. 6. The coarse-grained model illustrates the same effect of spatial variation in the drag and lift force. (A) The position of the RBC is shown at indicated times as it moves through the channel. (B and D) Time and initial condition averaged drag and lift force versus layer thickness and spatial frequency. (C and E) Time and initial condition averaged fraction of drag ($|\langle \text{Drag} \rangle|/(|\langle \text{Drag} \rangle| + |\langle \text{Lift} \rangle|)$) and lift force ($|\langle \text{Lift} \rangle|/(|\langle \text{Drag} \rangle| + |\langle \text{Lift} \rangle|)$) versus spatial frequency over all values of permeability. In all simulations, the thickness is fixed to be $h = 0.1827$. In panel B and D a 95% confidence intervals are plotted at each data point. Parameters are summarized in Table III.

initial conditions that uniformly sample one wavelength of the sine wave (Fig. S4B). Using the same set of permeabilities, we then calculated the resulting drag and lift. We find that increasing the spatial frequency only decreases the wall-induced migration of the RBC when the layer is highly permeable but does not affect its migration when the layer is nearly impermeable (Fig. 6, B-E). When exploring the influence of the ESL’s thickness, we find that the migration of the RBC is
significantly inhibited when a thick ESL is present and such a dependency persists for all values of ESL permeability (Fig. 7).

**FIG. 7.** The coarse-grained model illustrates the same effect of thickness in hindering the near-layer motion of the RBC. (A and C) Time and initial condition averaged drag and lift force versus layer thickness for different values of permeability. (B and D) Time and initial condition averaged fraction of drag \(\frac{|\langle \text{Drag} \rangle|}{|\langle \text{Drag} \rangle| + |\langle \text{Lift} \rangle|}\) and lift force \(\frac{|\langle \text{Lift} \rangle|}{|\langle \text{Drag} \rangle| + |\langle \text{Lift} \rangle|}\) versus thickness over all values of permeability. In all simulations, we assume the ESL is in a healthy condition corresponding to a spatial frequency of 1. In panel A and C a 95% confidence intervals are plotted at each data point. Parameters are summarized in Table III.

*Cell-free layer estimation for healthy and disrupted ESL*

The ESL plays an essential role in maintaining a healthy microcirculation due to its antithrombotic and anticoagulation functions. However, with infection or septic conditions, damage of the ESL occurs, enabling leukocyte and platelet adhesion and disrupt microcirculation [24, 43]. In
TABLE IV. Parameters for the whole blood simulations (Fig. 9)

| Parameter | Description       | Value               |
|-----------|-------------------|---------------------|
| Re        | Reynolds number   | 0.01 [25, 38]       |
| $k_s^{RBC}$ | Elastic spring constant | 3 $\mu$N/m [39] |
| $k_s^{Leuko}$ | Elastic spring constant | 30 $\mu$N/m [46] |
| $k_b^{RBC}$ | Bending constant  | $2 \times 10^{-19}$ N$\cdot$m [39] |
| $k_b^{Leuko}$ | Bending constant  | $2 \times 10^{-18}$ N$\cdot$m [47] |
| $k_a$      | Area preserving constant | 185 N/$\mu$m$^2$  |
| $k_{tether}$ | Tether force constant | 3200 N/$\mu$m  |
| $k_p$      | Porosity constant | 0 $\mu$m$^2$        |

recent years, new therapeutic approaches targeting the ESL have been proposed to improve microcirculation for sepsis patients [24]. Yet further work is needed to quantify the relationships between ESL disruption and microcirculatory function. To understand the changes in the microcirculation in vessels with healthy and disrupted ESL, we applied the coarse-grained model (see Materials and Methods) using ESLs extracted from medical images provided in [24] (see Fig. 3) using [37]. The number of RBCs in the healthy condition is estimated to be 48, and we assume these RBCs are placed uniformly across the region bounded by the healthy ESL. To determine the number of RBCs and their initial positions in the disrupted condition, we used the same initial set of cells as in the healthy condition and removed those ones that lie outside the region enclosed by the disrupted ESL.

We begin by considering a system that contains only RBCs. As an output of the model, we estimated the maximum and mean RBC velocity in the horizontal direction at steady state in both
cases. In the case of the disrupted ESL, we find that the blood flow decreases significantly with a 44% decrease in the maximum RBC velocity and a 39% decrease in the mean RBC velocity (Table V). To quantify the effect of the disrupted ESL on the microcirculation, we focus on the variation of the cell-free layer (CFL) thickness, which is an important parameter in several models of microvascular blood flow [5, 12, 48, 49] and has been shown to increase with increasing flow rate [50]. To determine the CFL thickness in each case, we averaged 800 vertical slices of the steady-state density plot (see Fig. 8, Video S1 and S2) evenly spaced from $x = 10$ to $x = 70$. In each slice, we measured the distance between the outer edge of the RBC core and the ESL, similarly to the CFL measurements done in experiments [9, 10]. In the healthy condition, we find that the top CFL is thicker than the bottom CFL. On the contrary, the difference in thickness between the top and the bottom CFL is small for the disrupted ESL (see Table V). Further comparing the thickness of top and bottom CFL, we find that the thickness of the CFL increases significantly in the disrupted condition while the RBC velocity decreases, demonstrating a negative correlation between the CFL thickness and blood velocity and the vessel diameter.

![Figure 8](image_url)

**FIG. 8.** The coarse-grained model shows that the cell free layer increases with disrupted ESL. (A and C) Schematics of the ESL layout (in red) and the positions of the RBCs at $T = 200$ with healthy (A) and disrupted ESL (C). (B and D) The steady-state density of RBC distribution. Each ESL is assumed impermeable ($k_p = 0$). RBC and ESL parameters are summarized in Table III.
To further investigate the influence of the ESL on the CFL in detail, we note that the plasma is a mixture of RBCs, leukocytes, and platelets. Therefore, we generalized the model to include leukocytes. Here leukocytes are modeled as circles of radius 6 µm. In the healthy case, two leukocytes are randomly placed within the blood vessel. The number of leukocytes are chosen so that there is at least one in the disrupted case and the relative ratio between leukocytes and RBCs remains close to that reported from experiments [51]. As before, we calculated the maximum and mean RBC velocity in the horizontal direction and estimated the thickness of the CFL at steady state in both the healthy and disrupted case (see Fig. 9, Video S3 and S4). We find that the RBC velocity decreases notably in the disrupted case (Table V) with a 51% decrease in the maximum RBC velocity and a 50% decrease in the mean RBC velocity. In comparison to the amount of decrease in the RBC velocities estimated from the RBC-only model, we find that the presence of leukocytes further slows down the motion of RBCs in the case of the disrupted ESL. Examining the resulting thickness of the CFL, as in the RBC-only model, the top and bottom CFL become significantly thicker when the ESL is disrupted. In both cases, the CFL is nonaxisymmetric.

The width of the CFL is influenced by the vessel diameter. The fraction of the vessel diameter occupied by the CFL in arterioles in the rat cremaster muscle was found to increase from 14% to 17% for vessel diameter ranging from 5 to 8 µm but to gradually decrease from 18% to 12% as the diameter increases from 8 to 25 µm [50]. As a first comparison, we followed the procedure in [50] and examined the relation between the fraction of mean CFL width over diameter and vessel diameter using the healthy and disrupted cases. For the RBC-only scenario, the fraction decreases from approximately 18% in the disrupted case of a mean vessel diameter of approximately 20µm to 7% in the healthy case of a mean vessel diameter of approximately 31µm. For the whole blood model, we again found that the fraction decreases as the vessel diameter is increased. In the disrupted condition (mean vessel diameter of 20µm), the fraction is estimated to be 13% and is decreased to 8% in the healthy condition (mean vessel diameter of 31µm).

We also compared the dimensionless thickness of the CFL, defined as the fraction of the CFL thickness over the vessel diameter, $R$, to modeling results given by [52] and experimental data provided by [8] extracted using [53]. In Fig. 10 we show the results of using our models against those from [8, 52] for discharge hematocrit level of 45% and 10%, corresponding to the healthy and disrupted case in our setup respectively. We see that the dimensionless thickness of the CFL estimated from the simulations using both the RBC-only and the whole blood model is in agreement.
FIG. 9. Whole blood simulation with extracted ESL. (A and D) Schematics of the ESL layout (in red) and positions of the RBCs and Leukocytes at $T = 200$ with healthy (A) and disrupted ESL (D). (B and E) The steady-state density of RBC distribution. (C and F) The steady-state density of Leukocyte (WBC) distribution. Each ESL is assumed impermeable (${k_p} = 0$). RBC, Leukocytes and ESL parameters are summarized in Table IV.

with the available observations to statistical error.

IV. DISCUSSION

The endothelial surface layer plays a critical role in regulating circulatory system. Under healthy conditions, the ESL acts as an antithrombotic and anti-inflammatory agent that prevents cells from sticking to the layer. Disturbed ESL, such as occurs during sepsis, can drastically change the blood flow and the dynamics of cells including the wall-induced migration. Using a combination of detailed and coarse-grained computational models via the immersed boundary method, we have examined
TABLE V. RBC velocity in the horizontal direction and estimated CFL thickness with extracted ESL

|                | Maximum RBC velocity [mm/s] | Mean RBC velocity [mm/s] | Top CFL [µm] | Bottom CFL [µm] |
|----------------|-----------------------------|---------------------------|--------------|----------------|
| RBC-only       |                             |                           |              |                |
| Healthy        | 0.708                       | 0.528                     | 3.316        | 1.101          |
| Disrupted      | 0.396                       | 0.324                     | 3.633        | 3.99           |
| Whole Blood    |                             |                           |              |                |
| Healthy        | 0.648                       | 0.444                     | 3.705        | 1.1            |
| Disrupted      | 0.264                       | 0.18                      | 3.289        | 1.896          |

FIG. 10. Dimensionless thickness of the CFL versus vessel diameter and discharge hematocrit. The black solid and dashed curves represent results extracted from [52]. Red solid squares are experimental data points taken from [8]. Error bars with a circle and an asterisk represent the simulation results in the healthy case for the RBC-only and the whole blood model respectively. Error bars with a diamond and a cross represent the simulation results in the disrupted case for the RBC-only and the whole blood model respectively. In all four cases, error bars correspond to 95% confidence intervals estimated from the simulation results.

The influence of the ESL on the wall-induced migration of the RBC. By simulating the wall-induced migration of a single RBC, we find that the layer thickness plays a dominant role in hindering the
RBC's ability of moving away from the layer and is independent of the ESL permeability. We note that when the blood vessel is lined with a thick ESL, it is analogous to a reduced vessel diameter. When the layer is impermeable, the ESL acts as a solid wall. Increasing the ESL thickness decreases the vessel diameter, resulting in a decrease in the blood flow and an increase in the resistance in the vicinity of the layer [7, 52, 54]. As a result, it then increases the amount of drag and hinders the migration of the RBC. A similar increasing trend in the drag is observed when the layer is highly permeable. In this case, the migration of the RBC is inhibited and is likely caused by a combined effect of ESL spatial variation as discussed previously in Results and a reduced vessel diameter due to the stiffness of ESLs. On the contrary, the layer’s spatial variation has a minimal effect in the healthy condition when the permeability is small. When the ESL is highly permeable, the RBC spends a longer time near the layer, causing the RBC to interact more with the ESL. As a result, the spatial variation of ESL has a more prominent effect on RBC migration in this case.

The amount of drag and lift force experienced by the RBCs together controls the formation of the CFL. Our key finding from using the medical images from [24] is that blood flow is significantly decreased when the ESL is disturbed during sepsis and that the presence of ESL contributes to a negative correlation between the CFL thickness and the vessel diameter. This underscores the potential important role played by the ESL in affecting the motion of RBCs and the formation of the CFL under various pathological conditions. We note that our whole blood simulations have revealed the non-axisymmetrical nature of the CFL in the healthy case, similar to that observed in experiments [9]. As we showed such non-axisymmetry is also preserved in the case when the ESL is disturbed. However, more comprehensive studies using ESLs during sepsis are required to draw an affirmative conclusion.

**2D versus 3D model.** While 3D simulations have become a standard approach in many context, performing long-time simulations in 3D at realistic hematocrit remains computationally challenging. On the other hand, 2D simulations are computationally feasible for long-time simulations, and allow us to gain insights that may be used as a foundation for performing 3D simulations more effectively in future work.

Another limitation of the 2D model is that it neglects the shear viscosity in the cell membrane, which is believed to impact dynamic deformations of the RBC [55–58]. Here we follow the approach used in previous studies [22, 23] in which the shear viscosity is neglected in modeling the membrane.

**Role of ESL in vessel function.** In this work, we take inspiration from the changes that occur
during sepsis to study the effects of various physical properties of the ESL and gain valuable insights on the effect of changes in the ESL surface structure arising during disease on blood flow and the distribution of RBCs in the vessel. Here the issues of ESL damage, change in permeability, and blood flow in a narrowed vessels are intimately coupled as the changes in vessel structure during sepsis is induced by leukocytes being trapped in the ESL. While the ESL has various other physiological roles e.g. interaction with platelets leading to coagulation that are outside the scope of this work, it would be a promising future direction to investigate the influence of a more comprehensive collection of the physiological properties of the ESL on vessel function.

Effect of ESL in other types of blood vessels. Here we have only discussed the influence of ESL topography in capillaries far away from the heart and blood flow is essentially steady and laminar. An interesting future direction would be to consider the effect of ESL in arteries, in which blood flow could occurs at higher Reynolds numbers, and to investigate how ESL controls the motion of RBCs and the formation of the CFL in the presence of pulsatile flow.

Lubrication effect. Additionally, in the present work we initialized the RBC at a position that is at least four grid points away from the layer so that lubrication effect can be neglected. Another interesting future direction would be to incorporate lubrication corrections within immersed boundary simulations to resolve the thin fluid layers that arise when cells come within subgrid distances of vessel walls [25].

V. AUTHOR CONTRIBUTIONS

Y.Z. and T.G.F. designed the research, performed the research, analyzed output, and wrote the article.

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VII. SUPPORTING CITATIONS

References [18, 20, 28, 36, 37, 59–61] appear in the Supporting Material.

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SUPPORTING MATERIAL

S1. Finite Difference approximation for spatial discretization

In what follows, we employ a modified staggered-grid spatial discretization for the equations of blood developed by [34]. We assume $\Omega$ is a rectangle of size $L_x$ by $L_y$ discretized into an $N_x \times N_y$ rectangular grid with grid spacing $\Delta x = L_x/N_x$ and $\Delta y = L_y/N_y$. The center of the $(i,j)^{th}$ rectangular grid cell is located at $x_{i,j} = ((i + \frac{1}{2}) \Delta x, (j + \frac{1}{2}) \Delta y)$, where $i = 0, \ldots, N_x - 1$ and $j = 0, \ldots, N_y - 1$. The pressure $p(x,t)$ is defined at the centers of the rectangular grid cells, and the values of $p$ on the grid are denoted by $p_{i,j}(t) = p(x_{i,j},t)$. We denote by $u_{i,j}(t)$ the fluid velocities at time $t$ and $f_{i,j}$ the external force of the $(i,j)^{th}$ grid cell and both are located at the bottom-left corner of the $(i,j)^{th}$ grid cell (see Fig. S3). The spatial differential operators in Eq. 1 are discretized using a second-order accurate scheme as in [25]

$$
\text{Re} \frac{d u_{i,j}}{dt} + (G p)_{i,j} = (L u)_{i,j} + f_{i,j} \tag{S1}
$$

The gradient of $p$ is approximated at the cell corner by

$$(G p)_{i,j} = \frac{1}{2} \begin{bmatrix}
\frac{p_{i,j} - p_{i-1,j} + p_{i,j-1} - p_{i-1,j-1}}{\Delta x} \\
\frac{p_{i,j} - p_{i,j+1} + p_{i-1,j} - p_{i-1,j+1}}{\Delta y}
\end{bmatrix}, \tag{S2}
$$

and the divergence of $u$ is approximated by

$$(D \cdot u)_{i,j} = \frac{1}{2} \left(\frac{u_{i+1,j} - u_{i,j} + u_{i+1,j+1} - u_{i,j+1}}{\Delta x} + \frac{v_{i,j+1} - v_{i,j} + v_{i+1,j+1} - v_{i+1,j}}{\Delta y}\right). \tag{S3}
$$

The Laplacian of $u$ is approximated via centered difference method as

$$(L u)_{i,j} = \begin{bmatrix}
\frac{(u_{i,j} - 2u_{i+1,j} + u_{i+2,j})}{\Delta x^2} \\
\frac{(v_{i,j} - 2v_{i+1,j} + v_{i+2,j})}{\Delta y^2}
\end{bmatrix}. \tag{S4}
$$

The object immersed in $\Omega$ is parametrized by the Lagrangian coordinate $q \in [0, L_q]$ and is discretized using $N_q$ points with grid spacing $\Delta q = L_q/N_q$. We denote by $X_k(t)$ the $k^{th}$ Lagrangian points in Eulerian coordinates and is defined as $X_k(t) = X(k \Delta q, t)$, $k = 0, 1, \ldots, N_q - 1$. To obtain the velocity of each immersed boundary point using Eqs. 12 and 13, we need the discrete delta function, $\delta_h(x)$, for interpolating and spreading. In the present work, we use the standard 4-point delta function, $\delta_h(x) = \frac{1}{\Delta x} \phi \left( \frac{x}{\Delta x} \right) \frac{1}{\Delta y} \phi \left( \frac{y}{\Delta y} \right)$ [28] and the resulting discretized immersed boundary
velocity is given by
\[
U_k(t) = \sum_{i=0}^{N_x-1} \sum_{j=0}^{N_y-1} u_{i,j}(t) \delta_h(x_{i,j} - X_k(t)) \Delta x \Delta y,
\] (S5)
and the external force at each Eulerian grid point is given by
\[
f_{i,j}(t) = \sum_{k=0}^{N_q-1} F_k(t) \delta_h(x_{i,j} - X_k(t)) \Delta q,
\] (S6)
where the Lagrangian force at the immersed boundary points, \( F_k \), is obtained using \( X_k(t) \) through energy functions (see Eqs. S9 – S11).

1. \textit{Implementing Tether Points}

The implementation of tether points introduce tether forces to blood flow. The discrete external force appeared in Eq. S1 is given by
\[
f_{i,j}(t) = f_{i,j}^{RBC}(t) + f_{i,j}^{tether}(t),
\] (S7)
where \( f_{i,j}^{RBC}(t) \) is interpolated from the Lagrangian force induced by the RBC and the ESL (Eq. 8) and \( f_{i,j}^{tether}(t) \) is the interpolated tether force using the Lagrangian tether force given by Eq. 10
\[
f_{i,j}^{tether}(t) = \sum_{l=0}^{N_l-1} F_{tether}^{l}(t) \delta_h(x_{i,j} - \hat{X}_l(t)) \Delta q,
\] (S8)

2. \textit{Imposing Physical Boundary Conditions}

The discrete spatial differential operators (Eqs. S2-S4) may require modifications near physical boundaries. To simplify the presentation, we restrict our attention to the domain boundary in the vicinity of the grid cell \((0,j), 0 \leq j \leq N_y-1\) that locates along the left side of \( \Omega \). On the staggered grid implemented in the present work, both \( u \) and \( v \) are specified on the boundaries of \( \Omega \). In this case, no boundary condition for the pressure is needed. As such no expressions are required for the ghost values \( p_{0,j-1}, u_{0,j-1} \) or \( v_{0,j-1} \).

The discrete spreading and interpolating operator \( \delta_h(x) \) also have to be modified near the domain boundary, in particular, when the support of \( \delta_h(x) \) overlaps with one boundary. One choice introduced in [60] is to modify \( \phi(r) \) and construction a new \( \delta_h(x) \) near the boundary. Here we chose another approach implemented in [61]. We use the standard 4-point delta function [28] for spreading and interpolating but extend the domain with sufficiently many ghost cells so that
the support of all delta functions is strictly within the extended domain. The ghost values of $u_{0,-1}$ and $v_{0,-1}$ are the mirror inversion of $u_{0,1}$ and $v_{0,1}$ respectively.

3. Discrete energy functions

The RBC immersed in $\Omega$ is discretized into $N_R$ points with spacing $\Delta q = L_R/N_R$, and the positions of Lagrangian points in Eulerian coordinates are given by $X_k(t) = X(k\Delta q, t)$, $k = 0, 1, \ldots, N_R - 1$. Discretizing the elastic energy for stretching/compressing (Eq. 5) we have

$$E_{\text{spring}} = \frac{k_s}{2} \sum_{i=0}^{N_R-1} \left( \frac{\|X_{i+1} - X_i\|}{\Delta q} - 1 \right)^2 \Delta q. \tag{S9}$$

Similarly, the discrete bending energy is given by

$$E_{\text{bend}} = \frac{k_b}{2} \sum_{i=0}^{N_R-1} \left( \frac{\|X_{i+1} - 2X_i + X_{i-1}\|}{\Delta q^2} \right)^2 \Delta q, \tag{S10}$$

and the discrete area-preserving penalty energy is

$$E_{\text{area}} = \frac{k_a}{2} \left( \sum_{i=0}^{N_R-1} \frac{\|X_i \times X_{i+1}\|}{2} - A_0 \right)^2, \tag{S11}$$

where indices are computed modulo $N_R$.

We assume that each ESL is discretized into $N_E$ Lagrangian points having spacing $\Delta \hat{q}$, and the positions of Lagrangian points in Eulerian coordinates are given by $\hat{X}_k(t) = X(k\Delta \hat{q}, t)$, $k = 0, 1, \ldots, N_E - 1$. In the detailed model, in which the ESL is described as a collection of fiber bundles, each fiber is discretized into Lagrangian points connected by elastic springs with rest length. The corresponding discrete elastic spring energy is identical to Eq. S9.

S2. Convergence study

As a simple control for convergence, we solve the unsteady Stoke equation (see Results) that governs the motion of the blood flow using the numerical method stated in SI Section VII S1 on a square domain of side length 1 with grid size that are subsequently halved.
1. Method of Manufactured Solution

To demonstrate the convergence of the method, we use the method of manufactured solution with \( \text{Re} \) set to be 0.01. We begin by considering the solution

\[
\mathbf{u} = \begin{bmatrix} u \\ v \end{bmatrix} = \begin{bmatrix} \cos(2\pi t) \sin(2\pi y) \\ 0 \end{bmatrix}, \tag{S12}
\]

with \( p = 0 \) the force, \( \mathbf{f} \), is given by

\[
\mathbf{f} = \begin{bmatrix} 2\pi \sin(2\pi y) (2\pi \cos(2\pi t) - 0.01 \sin(2\pi t)) \\ 0 \end{bmatrix}. \tag{S13}
\]

In Fig. S1A we plot the \( L^1 \) and \( L^\infty \) error between the approximated solution and the true solution, \( u_{\text{approx}} - u_{\text{true}} \), as the grid size is successively halved. In both cases, we observe a second-order convergence. To examine the convergence in a more complex scenario where the vertical velocity, \( v \), also changes in time and space, we consider another solution

\[
\mathbf{u} = \begin{bmatrix} (2y^3 - 3y^2 + y) \cos(2\pi(x-t)) \\ 2\pi(0.5y^4 - y^3 + 0.5y^2) \sin(2\pi(x-t)) \end{bmatrix}, \tag{S14}
\]

with \( p = 0 \) the force is given by

\[
\mathbf{f} = \begin{bmatrix} f_x(x,y) \\ f_y(x,y) \end{bmatrix}. \tag{S15}
\]

where

\[
f_x(x,y) = 2(2y-1)(2\pi^2y(y-1) - 3) \cos(2\pi(x-t)) + 0.02\pi y(2y^2 - 3y + 1) \sin(2\pi(x-t)), \tag{S16}
\]

and

\[
f_y(x,y) = -0.02\pi^2(y-1)^2y^2 \cos(2\pi(x-t)) + 4\pi^3(y-1)^2y^2 \sin(2\pi(x-t)) - 2\pi(1 - 6y + 6y^2) \sin(2\pi(x-t)). \tag{S17}
\]

We repeat the calculation of the \( L^1 \) and \( L^\infty \) error between the approximated solution and the true solution, \( u_{\text{approx}} - u_{\text{true}} \), as the grid size is successively halved. We observe a second-order convergence in both cases (Fig. S1B).

2. Flow over a Backward-Facing Step

To examine the accuracy of numerical methods on the Reynolds number, we simulated the flow over a backward-facing step in a channel and compare the reattachment length, \( x_r \), to those reported
FIG. S1. Convergence of the numerical approximation stated in SI Section VII S1 of the unsteady Stokes equation Eq. 1 with boundary conditions Eq. 2 to the true solution given by (A) Eq. S12 and (B) Eq. S14 with the corresponding force given by (A) Eq. S13 and (B) Eqs. S15–S17 as the mesh is refined. In both cases, the Reynolds number is fixed to be 0.01.

in [62–64] as the Reynolds number increases. Note, in order to be able to make a comparison, the full Navier-Stokes equation is used. For this benchmark problem, the domain is a rectangle of size \([0, 6] \times [0, 1]\) discretized into \(768 \times 128\) Cartesian (rectangular grid). The number of grid points in both \(x\) and \(y\) direction are chosen according to Fig. so that the result is fully resolved. A step of
height \( h_r = 0.5 \) is placed at the inlet. A parabolic flow of the form
\[
\mathbf{u} = \begin{bmatrix}
\max(24(1 - y)(y - 0.5), 0) \\
0
\end{bmatrix}
\]
is applied at the inlet with a zero Neumann boundary condition prescribed at the outlet. On the top and and the bottom wall of the channel, a zero Dirichlet boundary condition is used.

We estimated the reattachment length, \( x_r \), numerically as the Reynolds number is increased. In Fig. we show the estimated reattachment length over the height of the step, \( x_r/h_r \), for different Reynolds numbers in comparison to the experimental and computational results from [62–64]. The dependence of the reattachment length on Reynolds number is in good agreement with those reported in previous studies up to approximately \( \text{Re} = 250 \).

![FIG. S2. Reattachment length over the height of the step as a function of Reynolds number. Circles plot the data from [62]. Squares plot the data in [63]. Triangle plot the data reported in [64]. Solid red circles plot the result estimated from our simulations with the grid resolution fixed to be 768 × 128. Data provided in [62–64] are extracted using the online software [53].](image)

S3. Computational cost for the detailed and coarse-grained model

As a simple benchmark and comparison for the computational cost of the detailed and coarse-grained model, we fixed the thickness of the ESL to be 0.84μm and picked a relative extreme case
in which both the density of the bundles (detailed) and the spatial frequency (coarse-grained) are high. We examined three different values of $k_p = 0, 0.005$, and $0.01 \mu m^2$ with each case simulated till $T = 50$ s.

TABLE S1. Comparison of the computational cost of the detailed and coarse-grained model

| $k_p$       | Detailed | Coarse-grained |
|------------|----------|----------------|
| $0 \mu m^2$ | 5151 s   | 2842 s         |
| $0.005 \mu m^2$ | 5357 s   | 2844 s         |
| $0.01 \mu m^2$ | 5360 s   | 2849 s         |

To record the computational time for each case, we made use of MATLAB’s built-in timer `tic` and `toc`. For all three values of $k_p$, the coarse-grained model is approximately 2 times faster than the detailed model and the coarse-grained model becomes more computationally efficient as $k_p$ is increased (see Table. S1).
FIG. S3. An illustration of the staggered-grid spatial discretization in the present work. The fluid velocities $\mathbf{u} = (u, v)$ are defined in terms of those vector components that are normal to the edges of the grid cells, and the pressure $p$ is defined at the centers of the grid cells.
FIG. S4. Schematic of the ESL layout (in red) and the initial positions of the RBC used in simulations using (A) the detailed model and (B) the coarse-grained model.