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In Vitro Characterization of Lingering Red Blood Cells In Capillary Networks

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Introduction:

The distribution of red blood cells (RBCs) in the microvasculature is an important factor for the supply of the tissue with oxygen. It is known that the RBC distribution is spatially and temporally heterogeneous. One of the causes for the heterogenous distribution are lingering RBCs (LRBCs), i.e., RBCs which remain temporarily stuck at the apex of diverging bifurcations¹⁻³. However, the lingering of RBCs has not yet been fully characterized. Therefore, we investigated the behaviour and properties of LRBC in an in vitro model of a microvascular bifurcation.

Methods:

A PDMS microfluidic device featuring a symmetric bifurcation of microchannels mimicking the dimensions of capillaries (channel width 9.6 um) was perfused with rabbit RBCs suspended in a plasma-like solution $(\bar{u}_{RBC} \approx 0.68 \text{mm/s}, \text{Hematocrit} = 10 \%)^4$. An image sequence (Eclipse Ti-e, Nikon) of 4000 video frames of RBCs flowing through the bifurcation was recorded using a high-speed camera (395 frame/s, ORCA-Flash 4.0, Hamamatsu). The frames were pre-processed with a custom-written script⁴, and every RBC was then tracked by particle tracking velocimetry⁵ to determine position and velocity.

The transition time constant (τ) was used to discriminate between a LRBC and a non-lingering RBC (NLRBC) where τ is the time spent by the RBC in the bifurcation region divided by the mean time of all RBCs in this region. An RBC is considered lingering if it spends 50% more time in the bifurcation region than normal (LRBC τ >1.5 and NLRBCs τ <1.5).

Results:

In total, 378 bifurcation events were tracked. Out of these, 53 (14%) were classified as LRBC. Inspection of the lateral position of LRBC and NLRBC, indicated that LRBCs were flowing in the center of the parent vessel (PV, upstream of the bifurcation region), whereas the lateral position of NLRBCs was bimodally distributed with the two modes one eight of the channel width away from the centerline. Quantitative analysis confirmed that 96.2% of LRBCs were within one eighth from the centerline, whereas only 52.3% of the NLRBCs were in this region. After the bifurcation region, i.e. in the daughter vessels (DV), LRBCs were positioned closer to the inner wall (98.1% on the inner half width of the channel). In contrast, NLRBCs were more centered (28.9% on the inner half width). The mean RBC velocity in the DV was \bar{u}_{NLRBC} = 0.31mm/s for the NLRBC and \bar{u}_{LRBC} = 0.21mm/s for LRBCs. In the PV the difference between the LRBC and NLRBC was lower (\bar{u}_{LRBC} = 0.70mm/s, \bar{u}_{NLRBC} = 0.67mm/s).

Discussion:

In the PV, the only significant difference between LRBCs and NLRBCs was the lateral position. In the DV, the LRBCs were slower and stayed near the walls (as opposed to NLRBCs which were faster and more central). This different behaviour of LRBCs and NLRBCs is expected to result in a bifurcation bias in the next diverging bifurcation, which has been described previously as the history effect^{3,4}. Our results support the presence of the history effect and indicate LRBC should be taken into consideration when studying RBC distribution in capillary networks.

Participation

References

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