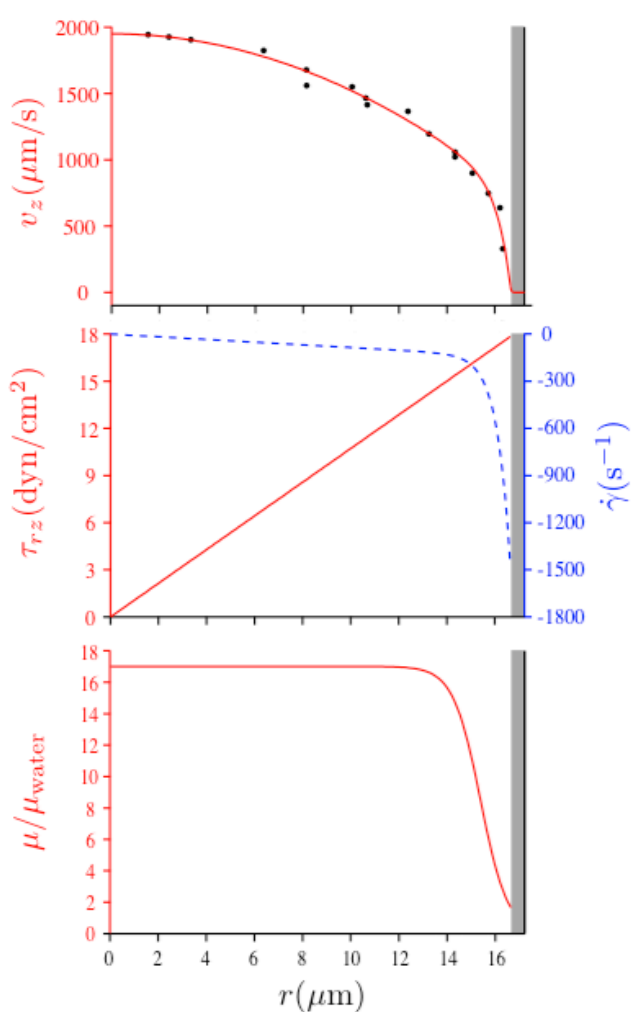


Microviscometric analysis in venules using μ -PIV

In order to significantly advance our current understanding of a great many important physiological and pathophysiological processes in cardiovascular research, it is becoming increasingly evident that a more sophisticated and realistic account of the underlying microhemofluidics involved in these processes will be required. In the microcirculation, vessel diameter, pressure gradient, and the volume fraction of red blood cells discharged by a vessel (discharge hematocrit) are among the most important determinants of a variety of flow parameters in a microvascular network, including the instantaneous red-cell fraction in the vessel (tube hematocrit), the relative apparent viscosity, the volume-flow rate, and the local shear stress. Owing to the significant heterogeneity in red-cell distributions that arise within microvascular networks *in vivo*, there typically exists a broad range of values the discharge hematocrit can assume for the many vessels in a given network, and even a significant variability in its value for a particular vessel in the network over time. This heterogeneity, coupled with the inability to accurately measure pressure gradients or discharge hematocrits *in vivo*, has confounded attempts at quantitatively characterizing blood flow in the microcirculation.

We have developed an extensive array of novel analytical tools that uses fluorescent micro-particle image velocimetry (μ -PIV) data to characterize microhemofluidics in post-capillary venules *in vivo* with an accuracy and detail that is unprecedented in microvascular research. This microviscometric method relies upon the assumption that the rheological properties associated with a particulate red blood cell suspension can be approximated by a continuous viscous fluid having a spatially varying viscosity. As our results have confirmed, this assumption turns out to be quite robust in microvessels and glass capillary tubes as small as 30 μm in diameter. Invoking this assumption, we have derived simple analytical expressions for the viscosity and hematocrit profiles over the vessel cross section that satisfy the conservation principles of mass and momentum and depend upon the experimentally obtained velocity distribution from our μ -PIV data. Once obtained, these profiles provide a wealth of information about dynamic, kinematic, and rheological properties of the flow (see figure 2).



$$\begin{aligned}\tau_{rz} &= \mu(r) \dot{\gamma}(r) \\ &= \frac{r}{a} \mu_a \dot{\gamma}(a), \quad 0 < r < a\end{aligned}$$

$$\frac{\mu(r)}{\mu_a} = \frac{r \dot{\gamma}(a)}{a \dot{\gamma}(r)}, \quad 0 < r < a$$

$2R$ (μm)	$R - a$ (μm)	$-dp/dz$ (dyn cm^{-3})	η_{rel}	$\dot{\gamma}(a)/\dot{\gamma}_P(R)$
34.5	0.56	18,895	5.15	6.2

FIGURE 2: Results from a 34.5- μm -diameter venule in the mouse cremaster muscle. (Top) Intravital fluorescent μ -PIV data (symbols) and predicted axisymmetric velocity profile extracted from the μ -PIV data. (Middle) predicted distributions in shear-rate, $\dot{\gamma} = dv_{fit}/dr$ (dashed blue, right axis), and shear-stress, τ_{rz} (solid red, left axis), whereby τ_{rz} was obtained from the expression above, where a is the radial coordinate location of the luminal interface of the glycocalyx and μ_a is the dynamic viscosity of blood plasma. (Bottom) Predicted viscosity profile, $\mu(r)$, obtained from the expression above and normalized with respect to water viscosity at the animal's body temperature. The shaded region near the vessel wall shows the predicted thickness of the glycocalyx. Tabulated values shown below the distributions include estimates of the glycocalyx thickness, $R - a$, the relative apparent viscosity, η_{rel} , and the ratio, $\dot{\gamma}(a)/\dot{\gamma}_P(R)$, of the interfacial shear rate, $\dot{\gamma}(a)$, to the shear rate, $\dot{\gamma}_P(R)$, corresponding to a Poiseuille flow in a tube of radius R having a centerline velocity corresponding to that predicted by the fit to the μ -PIV data. The viscosity distribution, $\mu(r)$, reflects the time-averaged viscosity at any point in the vessel cross section, and as such, is predicted to equal plasma viscosity only at the luminal interface of the glycocalyx (at $r = a$). Even in an approximately steady flow, experimental observations have shown that red cells transiently invade the plasma-rich region of microvessels, and contribute to the local viscosity there.

In particular, we have verified our ability to predict the pressure gradient, relative apparent viscosity, and tube and discharge hematocrits through comparison with directly measured quantities in glass capillary tubes 30 to 50 μm in diameter. In addition, local shear rate and local shear stress can also be estimated anywhere over the vessel cross-section. Furthermore, since our approach requires only systemic injection of a dilute suspension of commonly available microspheres, it can easily be implemented during the course of most intravital experimental protocols in a minimally invasive way.

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