

Micro-BioRheology

Macromolecular Dynamics in an Optimized Microfluidic Cross-Slot Geometry: Application to Biofluid Analogues and Prosthetic Fluid Formulation

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Viscoelastic fluids are a class of materials that display characteristics somewhere between those of solids (elasticity) and those of liquids (viscosity). Familiar examples of such fluids include melted cheese, melted plastic, sticky glue, saliva and mucus. The stretchy and stringy properties of these substances results from the presence of long and flexible molecules (either proteins or synthetic polymer molecules) that resist deformation under an applied elongational stress. The principal aim of this research project was to develop a microfluidic platform (a miniature flow channel) that could be used to characterize the properties of viscoelastic liquids undergoing stretching deformations. The microfluidic device would then be used to develop a more complete understanding of the physical flow properties (or “rheology”) of some important biological molecules, principally hyaluronic acid (HA), which is a polysaccharide found in large quantities in the synovial fluid (SF) that lubricates and protects the cartilage surfaces in mammalian joints. The strongly viscoelastic SF has been shown to become significantly degraded during the progression of degenerative joint diseases such as osteoarthritis (OA). Intra-articular injection of viscoelastic HA solutions are often used as a treatment for pain and discomfort in such cases, particularly in the knees where the SF is subjected to repeated cycles of stretching /compressing deformations during locomotion. An improved understanding of the rheology of HA solutions will aid the development of improved prosthetic fluids used for therapy. An ultimate aim is to develop a sufficiently sensitive characterization technique for HA that could provide a novel diagnostic method for joint diseases.

A microfluidic extensional flow device based on the principle of mutually bisecting channels (known as cross-slots) was fabricated according to a computer-generated optimized design, see Fig. 1(a).^{1,2} A comprehensive set of experiments was carried out in order to characterize the device and confirm that the flow conformed closely to the expectations based on the results of computational fluid dynamics (CFD) simulations. These experiments included the use of simple Newtonian fluids (like water) as well as “model” viscoelastic fluids composed of well-characterized polymer solutions. With polymer solutions, as the total flow rate through the Optimized-Shape Cross-slot Extensional Rheometer (OSCER) device is increased the deformation applied to the individual molecules can be seen using polarized light microscopy. As shown in Fig. 1(b), the polymer deformation is localized on the out-flowing axis of the OSCER device, which is where theoretical considerations and computer simulations also suggest the stress should be greatest, Fig. 1(c). Measurements of the intensity of the transmitted light in this region have been shown to be proportional to the fluid stress and can thus be used to obtain a measure of the resistance of the fluid to the imposed stretching or extensional deformation. This can also be correlated with the magnitude of the pressure loss with flow rate through the OSCER device.

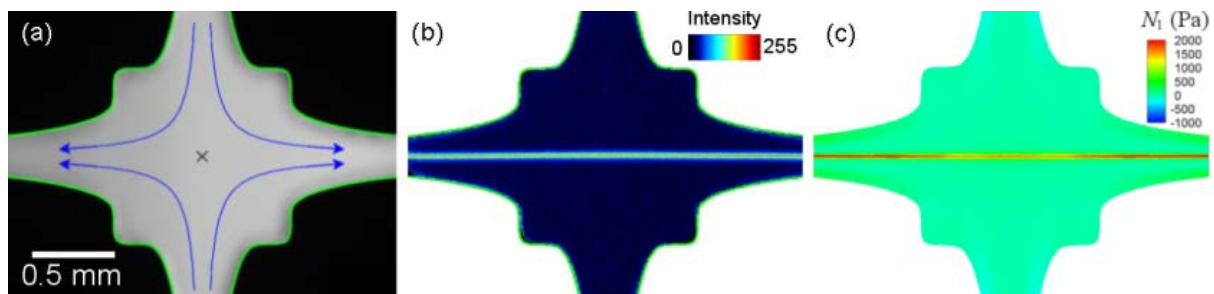


FIG 1: (a) Optical micrograph of the OSCER device with the computer-generated shape superimposed in green. The blue arrows indicate the flow direction. Stretching flow is generated along the horizontal axis of symmetry. (b) Polarized light micrograph obtained with a dilute polymer solution flowing in the OSCER device. (c) Computer-simulated first-normal stress distribution for the polymer solution.

Following the careful characterization of the OSCER device using model fluids, the techniques developed were applied to examine the extensional properties of viscoelastic HA solutions. Initially it was discovered that distinctive flow instabilities could develop in the OSCER device if the flow was driven beyond a certain critical rate that depended upon the fluid's viscoelastic properties. Accordingly, a detailed study was performed in order to ascertain the operating range of the extensional flow device, see Fig. 2(a-c).³ Understanding the causes of such flow instabilities is of great relevance to any polymer processing application and is also of fundamental interest among the physics and engineering communities. Within the stable operating space of the OSCER device, Fig. 2(c), the extensional viscosity of model synovial fluids based on hyaluronic acid solutions could be measured effectively, as shown in Fig. 2(d).⁴ For each fluid, the measured extensional viscosity is up to 50 times the shear viscosity measured in a conventional rheometer. This shows that the fluids are strongly “strain-hardening”. We believe this property may be extremely important in protecting joints such as the knees from sudden high-load impacts that occur during running and jumping, for example. The synovial fluid thickens as it is compressed between the joint surfaces, thus providing shock-damping and preventing damaging contact between the cartilage layers.⁴

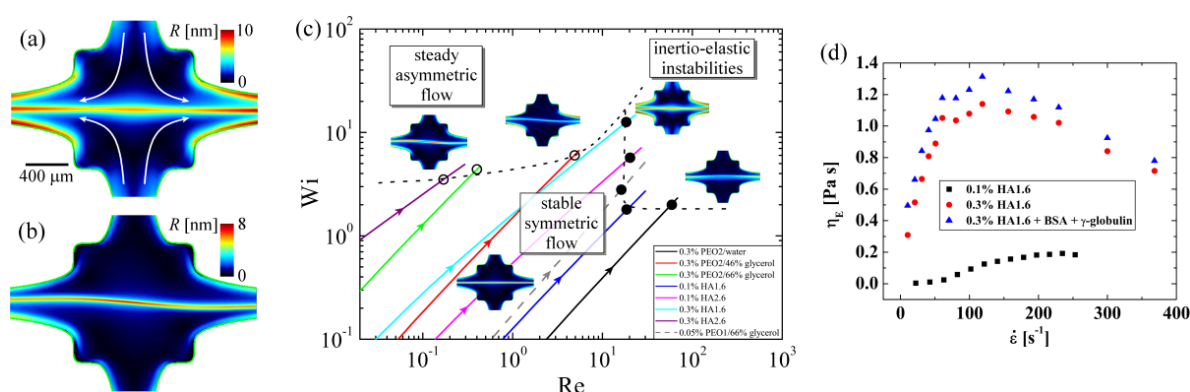


FIG 2: (a) “Inertio-elastic” flow instability for a low molecular weight HA solution. (b) Asymmetric “purely-elastic” flow instability in a high molecular weight HA solution. (c) Operating diagram for the OSCER device – within the region labeled “stable symmetric flow” the device functions as an extensional rheometer. (d) Extensional viscosity of various HA solutions measured using the OSCER device.

Further detailed experimentation on extensional flows of HA solutions has provided and confirmed some of the fundamental physical properties of the polysaccharide, such as the relationship between the relaxation time of the HA and its molecular weight and the refractive index difference of fully-oriented HA molecules.⁵ Combining this knowledge allows a semi-quantitative characterization of the distribution of molecular weight species present in an HA sample as well as an assessment of the HA concentration using extensional flow measurements. Thus, it may be possible to characterize the HA present in real synovial fluid in order to ascertain the health of the fluid or to determine if the fluid has become degraded.^{5,6} In future work, the methods developed show potential to be refined into a rapid microfluidic diagnostic technique based on extensional flow measurements with biopsy-sized samples of synovial fluid. This also promises to provide a useful toolbox for tracking the changes in synovial fluid hyaluronic acid composition, which should point to novel therapies and provide deep insights into the progression of joint disease.

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