BE/APh 161: Physical Biology of the Cell, Winter 2019
Homework #9
Due at the start of lecture, 2:30 PM, March 13, 2019.

Problem 9.1 (Stress softening of actin networks and buckling, 15 pts)**.**
In the Chaudhuri, Parekh, and Fletcher paper we discussed in lecture, stress softening occurred in actin filaments with a stress of about 230 Pa. The authors of that paper claim that stress softening occurs because actin filaments are buckling. Does this make sense to you? Perform an estimate to check.

Problem 9.2 (Optical cell stretching, 70 pts)**.**
We briefly discussed optical cell stretchers in lecture. Optical cell stretchers work by taking advantage of the difference in index of refraction between a cell and the surrounding solution to trap a free cell in two counter-propagating laser beams. The power of the laser is then increased to exert stress and elongate the trapped cell. The induced stress is proportional to the laser power. The constant of proportionality, F_G is dependent on geometry and cannot be ascertained. The deformation (strain) is measured by taking images with a light microscope. The process is illustrated in Figure 1. In this way, the mechanical properties of an entire cell can be measured.

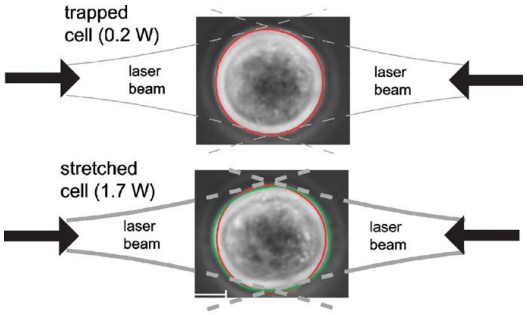

Figure 1: Schematic of an optical stretcher. The cell stretches along the axis parallel to the laser beams. The strain is given by the fractional change of the diameter of the cell along the stretching axis. Figure take from Wottawah, et al., *Acta Biomaterialia*, **1**, 263–271, 2005.

This technique was used to assess the mechanical properties of two mammalian cell types, 3T3 and SVT2 (which have reduced actin), in Wottawah, et al., *PRL*, **94**, 098103, 2005. In this work, the authors performed a stress step experiment in which a constant stress σ_0 was applied at $t = 0$, as in lecture. The stress was set back to zero at time $t = t_1$. The authors can obtain the creep compliance from this measurement.

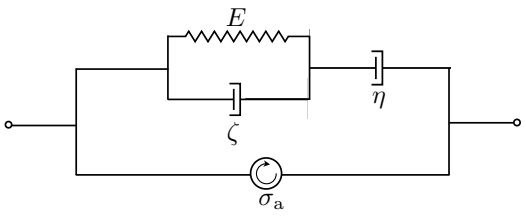

Figure 2: Schematic of an active Jeffreys fluid.

a) Derive an expression for the strain in the stress step experiment if we model the cell as an active Jeffreys fluid as in Figure 2. The stress step can be described mathematically as

$$\sigma(t) = F_G\, \sigma_0\, \theta(t)\, \theta(t_1 - t), \tag{9.1}$$

where $\theta(t)$ is the Heaviside step function. Assume the active stress is constant, given by σ_a.

b) The authors perform curve fits of the expression you derived in part (a) to get values for the parameters of the cell. Explain why they cannot independently measure E, η, and ζ, but only products thereof. Can a constant active stress be detected in this experiment?

c) The authors then use the curve fit parameters to compute the storage and loss moduli (E' and E'') of the cell. Derive expressions for the storage and loss moduli from the fit parameters. (*Note*: These reported storage and loss moduli are dependent on choosing a model for the viscoelastic behavior of the cell. This is not ideal, but is apparently a necessity due to experimental constraints.)

Problem 9.3 (Actin and gravity, 15 pts)**.**
The eggs of the frog *Xenopus* are very large single cells, measuring over a millimeter in diameter. The egg has a very large nucleus, measuring about 450 µm in diameter. Unlike nuclei many other eukaryotic cells, these giant nuclei have an actin network inside of them. Feric and Brangwynne (*Nat. Cell. Biol.*, **15**, 1253–1259, 2013) were studied this actin network with microrheology and then perturbed the network by treating the nucleus with the actin depolymerizing drug Latrunculin A.

a) Fig. 3 shows the tracks of beads of various radius that were injected into wild type nuclei of *Xenopus* eggs. Speculate on why the traces have the features that they do.

b) In wild type oocytes, the nucleus has thousands droplets of RNA and protein, called ribonucleoprotein bodies (RNPs), many of which are one micron in diameter or bigger, scattered throughout the nucleoplasm of the nucleus. When they treated the oocytes with Latrunculin A and left them overnight, Feric and

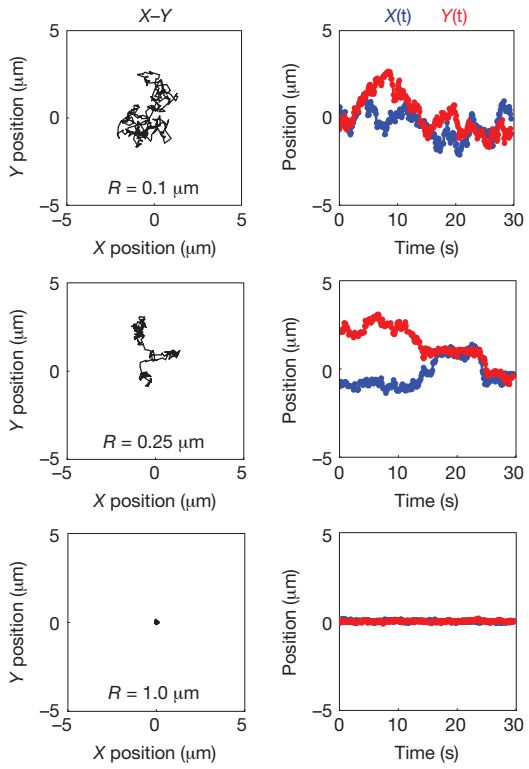

Figure 3: Tracks of beads of various size injected into the nucleus of a *Xenopus* oocyte. At left is a plot of the trajectory in the x-$y$plan and at right is the same trajectory where the x and y positions are plotted against time. Figure take from Feric and Brangwynne, *Nat. Cell. Biol.*, **15**, 1253–1259, 2013.

Brangwynne made a startling observation. In the morning, nearly all of the RNPs had fused together and were sitting at the bottom of the nucleus, apparently having sunk there. They determined that the density difference between the RNPs and the nucleoplasm was about 0.035 g/mL. (They determined this with clever experimentation; you should read the paper to see how they did it.) So far, we have been neglecting gravity because cells and the components inside them are small (obviously gravity affects larger scale organisms, like whole humans). How big do the RNPs need to be such that we need to start taking gravity into account? How would your answer change for a more typical nuclear size of about 10 μm?

c) Given these experimental results, is the wild type nucleoplasm more like an elastic of viscous medium?