For homework you have examined the concept of using a sandwich of two disks of radius R separated by a distance 2a in the z direction and which are oscillated in the θ direction with motion $θ = Δθ_0 \sin(ωt)$. (The geometry looks a lot like an Oreo with a wall or well around the rim and fluid in the center rather than that white stuff…). The inertia of the fluid causes the fluid to experience some shear in the θ-z plane, and centrifugal forces resulting from this non-uniform θ velocity lead to a radial inertial secondary current. In this project I want you to evaluate the possibility of using this geometry to do a separation of RBC’s infected with the malaria parasite. A recent article describing the problems associated with the detection process is Pootschi, et al., Trans Research 194:36-55, 2018. A key problem is that it is hard to visually identify the infected cells that are greatly outnumbered by the uninfected ones. The idea I would like you to explore is the possibility of separating infected cells from uninfected cells, preferentially concentrating them in the center of the disk for either direct imaging or further processing.

The proposed idea is to prepare a sandwich by removing the top plate and filling it with an isotonic buffer solution (isotonic means that it has the same osmotic pressure as the contents of the cells, preventing distortion of RBC’s) of chosen density. A drop of blood would be added to the center of the sandwich, and then the top disk would be put into place. The sandwich would be clamped, and then subject to the oscillatory rotation for a fixed length of time. Since the motion is just solid body rotation (albeit oscillatory), a whole stack of such plates could be oscillated at once for parallel processing. At the end of the oscillation time, infected cells (possibly along with the leukocytes, although it would be nice to get rid of these too) would be concentrated in the center and the uninfected RBC’s (that tend to obscure your view) would have been convected radially outwards. You then look at the cells near the center to determine if a malarial infection is present.

Separation in the radial direction can occur only if cells are separated to different locations in the z-direction. Migration in the z-direction occurs due to a combination of the θ-direction shear flow and the following mechanisms:

1) Sedimentation. Depending on the density of the buffer, cells will experience a vertical velocity due to Stokes sedimentation. The density of infected cells is less than that of uninfected cells. McAlister & Gordon (J. Parasitology, 62:664-669, 1976) report that schizont stage cells have a density of less than 1.043 g/cm³, trophozoite stage cells have a density between 1.081 and 1.091, while uninfected and slightly infected cells have a density greater than 1.091. Leukocytes have a density of 1.08 and RBC’s have a density of 1.11 (typically). Thus, there is an overlap between leukocytes and trophozoite infected cells.

2) Inertial migration. Cells (or spheres in general) will migrate in a channel flow due to inertial forces. This is the classic Segre-Silberberg effect, where particles will tend to congregate about 2/3 of the distance between the center and the walls. The migration velocity (for rigid spheres, anyway) was solved by Ho & Leal (J Fluid Mech, 65:365-400, 1974). It is reasonable to model the inertial migration process of a cell as a rigid sphere.
Note that this migration process depends on the size of the cell, where leukocytes are typically 11-15µm in diameter, while erythrocytes are disks about 7µm in diameter, but the effective spherical diameter would be a bit smaller. Thus, inertia will play more of a role for leukocytes than erythrocytes.

3) Deformation induced migration. In a channel flow a deformable drop or particle will migrate to the centerline (for low viscosity drops) or to an intermediate position for drops of viscosities similar to the suspending fluid. This was solved by Chan & Leal (1979). A cell isn’t really a drop, however, as it is more of an elastic capsule. These are also known to migrate to the centerline in a channel flow. This is still an area of active research. A very recent article (which also provides other useful references) of the combined effects of inertia and deformation is give by Alghalibi, et al., Phys Rev Fluids 4, 104201, 2019. An article which reports experimental measurements of focusing in oscillatory microfluidic flows is Mutlu, et al, PNAS 115:7682-7687 (2018). These references might be a good starting point to determine a model for cell migration in the proposed geometry. A bit of literature search is required here, and I will be interested to see what you come up with!

4) Shear-induced migration. Cell-cell interactions resulting from the shear flow will cause dispersion at moderate concentrations. This will cause a smearing out of the trajectories of individual cells. The diffusion coefficient would be proportional to the shear rate and the square of the particle radius, and would be (likely) proportional to the concentration in the dilute limit. It is reasonable to use the diffusivity determined in King & Leighton (Phys Fluids, 2001) for a dilute emulsion as an estimate of this effect.

5) Convection. In addition to the oscillatory velocity in the θ direction, there is an inertial secondary current in the radial direction (which you have calculated!) and a velocity in the z-direction which you can obtain from the radial velocity and the continuity equation. This will further affect the motion of the cells, as the other migration processes would be in addition to this convective velocity.

Given all this, what I propose for you to do for the final project is the following:

1) Calculate the velocity in the z-direction as well as the shear rate profile from your perturbation solution. Get the streamfunction in the r-z plane (it is proportional to the z-velocity) and plot up the fluid velocity contours in this plane.

2) Develop a simulation which tracks out the motion of the cells in this geometry. You can look at a case where the outer end is open in which cells which are convected out do not return (this could be accomplished by having a well at the outer edge, for example) or which is closed where the cells convected outwards would be returned on the same streamline at the outer edge (e.g., at the same value of the streamfunction), but in the mirror location. If you reflect things properly at the outer edge, and have no z-direction migration (e.g., sedimentation effects) included, a uniform initial concentration should stay uniform. This is a useful test of your simulation.

3) Add in the mechanism of sedimentation to your simulation, and determine conditions under which the desired separation would occur. Note that because the flow field is symmetric, you pretty much have to choose a buffer density which matches that
of the infected RBC’s, and even then neutrally buoyant particles don’t concentrate from this mechanism, they just flow around with the fluid. It is the ones which are not neutrally buoyant which will move radially outwards over time. Determine optimal parameters for doing the separation in a reasonable amount of time. Note that the frequency can’t be arbitrarily high, and the maximum shear rate in the system cannot exceed 1000s⁻¹ to avoid cell lysis. A reasonable gap width would be 200µm, and radius could be 2cm. A drop of blood is about 0.05cm³, so when compressed to 200µm it would occupy about the central 9mm radius of the disk. Note that cells can’t get closer than one radius from the top or bottom, and thus they would still experience some velocity in the radial direction. This is actually the basis of the sedimentation field flow fractionation technique, at least for pressure driven flow through a channel.

4) Determine a model for inertia and deformation induced migration from the literature sources and add them into your simulation to determine how these effects influence separation. This will likely speed things up substantially as the cells will move away from the walls and experience a higher velocity. It may also be possible to use these mechanisms to separate out the leucocytes which have the same density as the infected erythrocytes, and to actually collect the infected, neutrally buoyant cells in the center (the ultimate goal).

5) Add in the effect of finite concentration to see what shear-induced migration does to the separation. As a first step, you can use a diffusion coefficient which is characterized by some average particle concentration. This could be refined to yield a more accurate simulation.

Because of the nature of all of these drift mechanisms, as well as the radial velocity, only neutrally buoyant deformable cells of a particular size are likely to accumulate in the center (at least if the parameters are chosen correctly, anyway!). The key is to determine what parameters (if any) lead to this situation, and how long will it take for this separation to occur.

Note that the above are suggestions, not requirements: what I want you to do is to evaluate the proposed technology using the skills and concepts you have learned in this course and determine if there is any way it could lead to a practical separation to improve the detection of the malaria parasite in a drop of blood.

What to turn in: A written description of your equations, analysis, and conclusions, as well as any accompanying material such as graphs and computer programs.