## Force field calculation

The force field calculation used a reconstruction method to infer the force field from the previously measured displacement field

- 1. Input the Young modulus of the substrate you are using.
- 2. Choose a reconstruction method. Currently we have the choice between two detection methods:
  - 1. FTTC (Fourier-Transform Traction Cytometry): This method is the Matlab implementation of Benedikt Sabass (Sabass et. al, 2008 BiophysJ). This method uses L2-regularization.
  - 2. FastBEM (Fast Boundary Element Method): This method was implemented by Sangyoon J Han, summarized in Han et. al, 2015, Nature Methods. IMPORTANT: this function is computationally expensive. It will need more than 32GB to run.
- 3. Type in value for a regularization parameter.
- 4. Check **Use L-curve for regularization parameter selection** for objective parameter selection. Choose between L-corner vs. Optimal (depending on traction noise in a gel). If you select this option, the typed regularization parameter will be referenced for deciding the range of regularization parameter for building L-curve.
- 5. For FastBEM reconstruction method only
  - 1. Set the number of mesh points for the forward Solutions. It has to be a multiple of 2. *Flow will be calculated at only one time point for the whole movie*.
  - 2. Set a coefficient solving method: choose **backslash** for L2-regularization, or **1NormReg** for L1-regularization. (QR, svd, gsvd are mathematically the same as backslash. LapalcianReg and 1NormRegLaplacian are for L2-or L1-regularization with second derivative solution norm, respectively).
  - 3. Leave refinement parameter for L-curve calculation as 10. It is fine enough. The more this factor is, the more regularization parameters are tested in-between one order of magnitude (e.g. between 10<sup>-5</sup> and 10<sup>-6</sup>).
  - 4. If you already calculated the basis class functions, use **Select** to choose the location of the MAT field containing these basis class functions. If not, you can type in virtual file path there, then the basis class lookup table will

be saved in the path after it is calculated.

## 5. Click on **Apply**.

The output of force field calculation is a MAT file stored in a folder called "forceField". Clicking on **Result** will yield a pop-up window in which the reconstructed force field is overlaid onto the raw images. The traction magnitude map will be also created and can be viewed by clicking traction map radio button.