EE 367 Project Proposal
Three Dimensional Deconvolution Microscopy

Motivation

Fluorescence microscopy is a popular technique used to observe sub-cellular biological structures. When excited, each fluorophore emits light and this pattern is recorded by the microscope or the sensor. The issue with this technique is that even if a fluorophore is not in the focal plane of the system, it emits light and makes the image appear blurry. Deconvolution is a computational technique to remove the out-of-focus light from each image plane.

Project Overview

Deconvolution requires the knowledge of the optical point spread function (PSF), which describes how a single point is imaged through a system. There are many techniques to perform deconvolution both in 2 and 3 dimensions. I will focus on 3D Richardson Lucy deconvolution, which is an iterative method that assumes the presence of Poisson noise in the captured images. The fluorescence imaging problem can be written as:

$$\mathbf{A}\mathbf{x} = \mathbf{b}$$

In this particular case of 3D microscopy, $\mathbf{b}$ is the captured focal stack, $\mathbf{x}$ is the sample volume that is being imaged, and $\mathbf{A}$ is the circulant matrix that represents convolution in 3 dimensions. However, it is important to note that when these operations are carried out in the iterative updates, the matrix $\mathbf{A}$ is never created or calculated explicitly: all operations are done in the Fourier domain using the OTF (magnitude of the Fourier transform of the PSF) and the Fourier transform of the focal stack.

PSF Estimation

Richardson Lucy deconvolution is sensitive to the input PSF, and its accuracy affects how fast the algorithm converges, whether it converges at all, and whether it converges to the correct result. The first step for the project would be to work on estimating the correct PSF based on the parameters of the optical setup/microscope as well as accounting for any non-idealities in the system. Any aberrations would be accounted for by minimizing the difference between the theoretical PSF and the observed PSF. The minimization will be carried out over a set of Zernike polynomials that describe all possible aberrations in the system.

3D Richardson Lucy Deconvolution

Richardson Lucy deconvolution, as discussed in class, will be implemented in 3 dimensions. The important considerations will be making the code efficient enough that it can run through multiple iterations with large, 3D datasets. It will also be interesting to see whether certain 3D structures are better suited for deconvolution.

The iterative updates for the 3D case are the same as in the 2D case. Additional parameters to consider are the padding in the x,y, and z dimension in order to make the deconvolution process more efficient. The iterative update is as follows:

$$\mathbf{x}^{(q+1)} = \frac{\mathbf{A}^T(\frac{b}{\mathbf{A}x})}{\mathbf{A}^T\mathbf{1}}\mathbf{x}^{(q)}$$

3D Richardson Lucy Deconvolution with Total Variation Prior

Since fluorescence microscopy is mostly used for biological structures and individual fluorophores, we can assume that the gradients will be sparse. Therefore, adding a TV prior to the RL deconvolution algorithm can make the the deconvolve image stacks appear less noisy. One of the concerns with this method is losing detail information, and this will be explored further with the implementation of this technique. Another aspect of this algorithm is choosing the correct lambda value depending on the image stack that is being deconvolved, which can also be optimized.

In the 2D case, the anisotropic TV norm gradient is:

$$\nabla\lambda||\mathbf{Dx}||_1 = -\lambda(\frac{\mathbf{D_x x}}{|\mathbf{D_x x}|} + \frac{\mathbf{D_y x}}{|\mathbf{D_x x}|})$$

Since each dimension is treated independently in terms of minimizing the total variations, we simply add the gradient in the z direction for the 3D case:

$$\nabla\lambda||\mathbf{Dx}||_1 = -\lambda(\frac{\mathbf{D_x x}}{|\mathbf{D_x x}|} + \frac{\mathbf{D_y x}}{|\mathbf{D_y x}|} + \frac{\mathbf{D_z x}}{|\mathbf{D_z x}|})$$

Therefore, the iterative update for RL deconvolution in 3D with the TV prior would be:

$$\mathbf{x}^{(q+1)} = \frac{\mathbf{A}^T(\frac{b}{\mathbf{A}x})}{\mathbf{A}^T\mathbf{1} - \lambda(\frac{\mathbf{D_x x}}{|\mathbf{D_x x}|} + \frac{\mathbf{D_y x}}{|\mathbf{D_y x}|} + \frac{\mathbf{D_z x}}{|\mathbf{D_z x}|})}\mathbf{x}^{(q)}$$

Just like in the 2D case, it is possible for the denominator to either be 0 or the multiplicative update to be negative. Therefore, it is still essential to use small λ values and account for cases where $\mathbf{x}$ becomes negative.

Since we are adding sparsity in the z direction, it is important to keep in mind what kind of structures we are deconvolving. There will be a difference between volumes that only have objects in one plane versus samples that have objects scattered throughout. We can also add weighting factors to the gradients in each direction with prior knowledge of what the deconvolved sample should look like.

Alternating Direction Method of Multipliers and TV

With large datasets, RL deconvolution can be slow and take many iterations to converge to a solution. Implementing the deconvolution and Poisson denoising subproblems in the ADMM form can speed up the process as well as increase likelihood that the algorithm converges to the true solution.

The first step in modeling the deconvolution and denoising problem as ADMM would be to define the two objective functions:

$$\begin{aligned} &\text{minimize}_{\ x} \text{ - log } (p(\mathbf{b}|\mathbf{z_1})) + I_{\mathbf{R}+}(\mathbf{z_2}) \\ &\text{subject to } \begin{bmatrix}\mathbf{A}\\ \mathbf{I}\end{bmatrix}\mathbf{x} - \begin{bmatrix}\mathbf{z_1}\\ \mathbf{z_2}\end{bmatrix} = 0 \end{aligned}$$

The first term in the objective function corresponds to the MLE estimator for Poisson noise and the second term corresponds to the non-negativity constraint. The above can be solved iteratively with using proximal operators to update $\mathbf{x}, \mathbf{z_1}, \mathbf{z_2}$. Even though the iteration mechanism stays the same in 3D versus 2D, it is important to consider efficiency since the size of the variables grow considerably in the 3D case.

Adding in the TV prior would add three additional objective functions to the ADMM problem, one per direction in which total variation is to be minimized. The updated ADMM-TV problem would be:

$$\text{minimize}_{\,x} \text{ - log } (p(\mathbf{b}|\mathbf{z_1})) + I_{\mathbf{R}+}(\mathbf{z_2}) + \lambda||\mathbf{z}_3|| + \lambda||\mathbf{z}_4|| + \lambda||\mathbf{z}_5||$$
$$\text{subject to } \begin{bmatrix} \mathbf{A} \\ \mathbf{I} \\ \mathbf{D_x} \\ \mathbf{D_y} \\ \mathbf{D_z} \end{bmatrix} \mathbf{x} - \begin{bmatrix} \mathbf{z_1} \\ \mathbf{z_2} \\ \mathbf{z_3} \\ \mathbf{z_4} \\ \mathbf{z_5} \end{bmatrix} = 0$$

The above problem can be solved iteratively through the use of proximal operators. Compared to the 2D case, there is an additional variable, $\mathbf{z_5}$ to update.

ADMM with Different Priors

If there is time, I would also like to look into priors other than TV that could be applied for 3D deconvolution that work better with fluorescence microscopy data.

Timeline

- 2/5 • Tests and Simulation for PSF Estimation
- 2/12 • 3D Richardson Lucy deconvolution (implementation, test on real data)
- 2/19 • 2D RL + TV deconvolution (implementation, test on real data)
- 2/26 • 3D RL + TV deconvolution (implementation, test on real data)
- 2/26 • 2D ADMM + TV deconvolution (implementation, test on real data)
- 3/4 • 3D ADMM + TV deconvolution (implementation, test on real data)
- 3/9 • Final Presentation

References

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[3] Leon B Lucy. An iterative technique for the rectification of observed distributions. *The astronomical journal*, 79:745, 1974.

[4] William Hadley Richardson. Bayesian-based iterative method of image restoration*. *JOSA*, 62(1):55–59, 1972.