

Lab 9. (case) 3D deconvolution of the microscopy fluorescence image

Materials

- Five '*.mat' files
 - simSphere1.mat - a non-blurred microsphere
 - PSFG&Lsmall.mat - a calculated PSF data
 - simblurNoNoise.mat - a calculated blurred microsphere without noise
- Realdat.mat - a measured microsphere data
- PSFreal.mat - a measured PSF data

Tasks

1. Generate noise disturbed data using calculated blurred microsphere data;
 - Be careful, the original calculated blurred microsphere data assumes that the fluorescence intensity of each point is 1 and this fluorescence intensity should be adjusted to ensure the final value of image pixels are not exceed 1.
 - It is recommended to add background noise to simulate the real detector system.
2. Implement the inverse filtering and Wiener filtering method;
 - MATLAB function 'deconvwnr' is not allowed.
3. Implement the R-L iteration algorithm. (Optional)

Tasks(cont.)

4. Compare the inverse filtering, Wiener filtering and R-L iteration on simulated data;
 - the R-L iteration method can applied based on either of TASK 3 and MATLAB function 'deconvlucy';
 - try to change the noise model and SNR to compare and discuss the different results.
5. Apply the inverse filtering, Wiener filtering and R-L iteration on real data and compare the results.

Methods and Tips

Inverse filtering

$$\bar{F}(u, v) = F(u, v) + \frac{N(u, v)}{H(u, v)}$$

$$\bar{f}(x, y, z) = \mathcal{F}^{-1} \left\{ \frac{G(\omega_x, \omega_y, \omega_z)}{H(\omega_x, \omega_y, \omega_z)} \right\}$$

Weiner filtering

$$\bar{F}(u, v) = \left[\frac{1}{H(u, v)} \frac{|H(u, v)|^2}{|H(u, v)|^2 + \frac{S_\eta(u, v)}{S_f(u, v)}} \right] G(u, v) = \left[\frac{1}{H(u, v)} \frac{|H(u, v)|^2}{|H(u, v)|^2 + K} \right] G(u, v)$$

The inverse filter in Weiner filtering

$$\hat{H}(\omega_x, \omega_y, \omega_z) = \frac{H^*(\omega_x, \omega_y, \omega_z)}{\left(|H(\omega_x, \omega_y, \omega_z)|^2 + \frac{S_\eta(\omega_x, \omega_y, \omega_z)}{S_f(\omega_x, \omega_y, \omega_z)} \right)} \quad \text{NSR}$$

Tips

- $S_f(\omega_x, \omega_y, \omega_z)$
 - Power of the **non-blurred signal**
- $S_\eta(\omega_x, \omega_y, \omega_z)$
 - Power of the noise
- When $\text{NSR} = 0$
 - Inverse filtering = Wiener filtering

$$\frac{S_\eta(\omega_x, \omega_y, \omega_z)}{S_f(\omega_x, \omega_y, \omega_z)}$$

Tips

- For simulated / calculated data
 - Be careful, the original calculated blurred microsphere data assumes that the fluorescence intensity of each point is 1 and this fluorescence intensity should be adjusted to ensure the final value of image pixels does not exceed 1.
 - It is recommended to add background noise to simulate the real detector system.
 - $\text{blurred} * \text{factor} + \text{background noise} < 1$
 - Power of (non-blurred signal * **factor**)
- For real data
 - Power of non-blurred signal?

Tips

$$\hat{H}(\omega_x, \omega_y, \omega_z) = \frac{H^*(\omega_x, \omega_y, \omega_z)}{\left(|H(\omega_x, \omega_y, \omega_z)|^2 + \frac{S_\eta(\omega_x, \omega_y, \omega_z)}{S_f(\omega_x, \omega_y, \omega_z)} \right)}$$

- Avoiding extremely small numbers in the denominator
 - Set these numbers to be numbers a little bigger
 - Useful Matlab functions: eps, realmin, ...
 - eps = 2.2204e-16
 - sqrt(eps) = 1.4901e-08
 - realmin = 2.2251e-308

Maximize the likelihood

- Maximize the log likelihood of the Poisson process

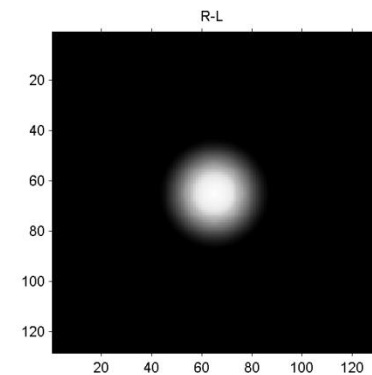
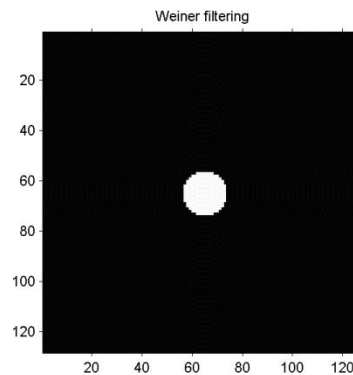
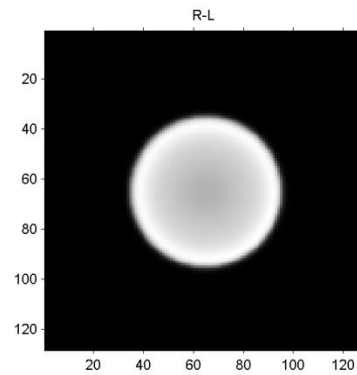
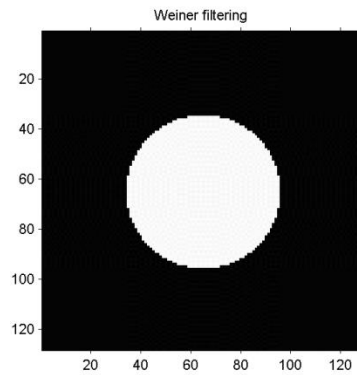
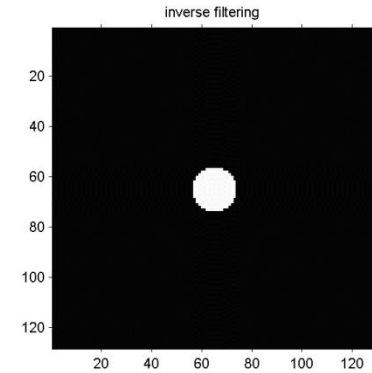
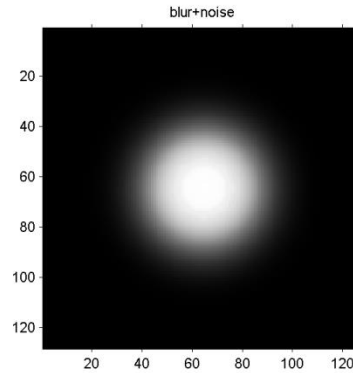
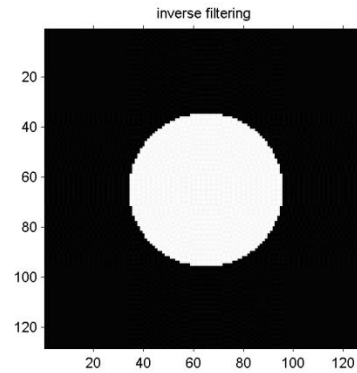
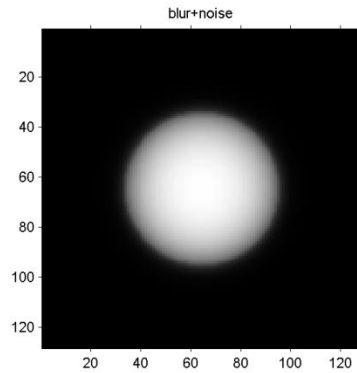
$$\max J(f) = \sum_{x,y,z} g * \log \lambda - \lambda$$

- Apply the gradient-based iterative search algorithm(R-L iteration algorithm)

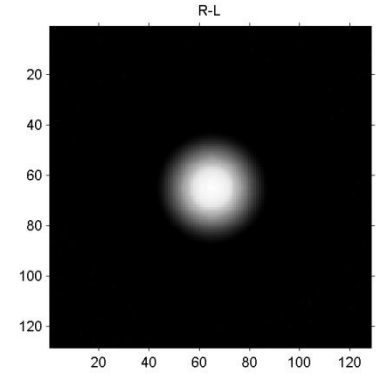
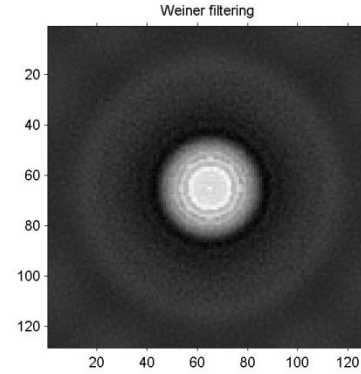
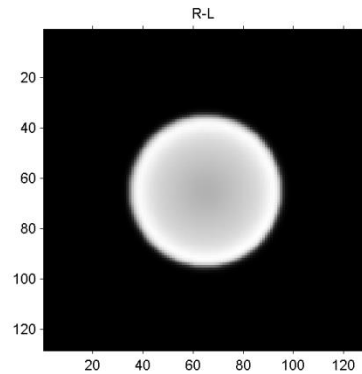
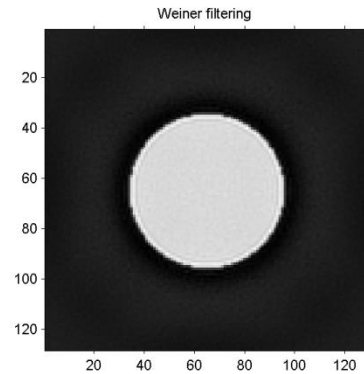
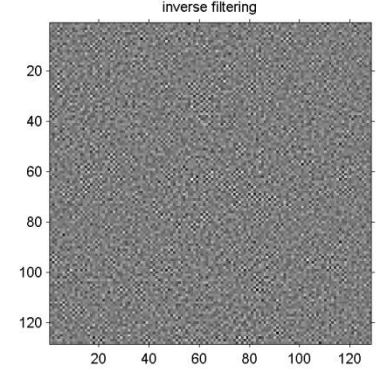
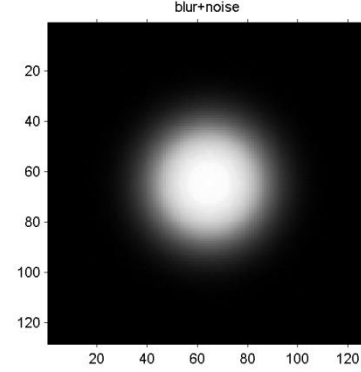
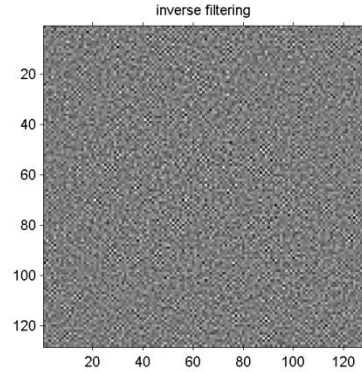
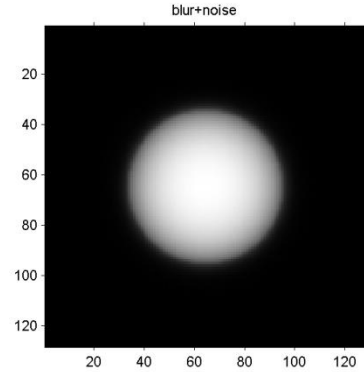
$$\bar{f}_{k+1} = \left[\frac{g}{\bar{f}_k \otimes h} \otimes (-h) \right] \bar{f}_k, \quad (-h) = h(-x, -y, -z)$$

- `imnoise`
 - Add noise to image
- `psf2otf`
 - Convert point-spread function to optical transfer function
- `fftn, ifftn`
 - n-D fft, ifft
- `flipdim`
- `deconvlucy`

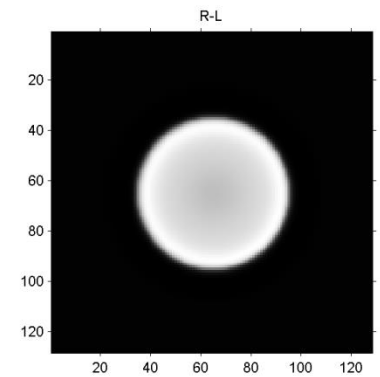
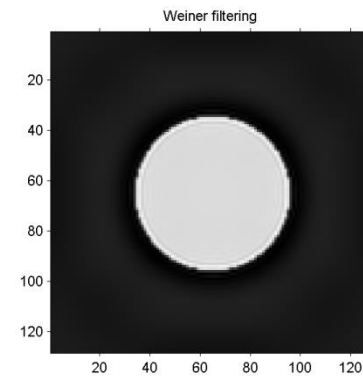
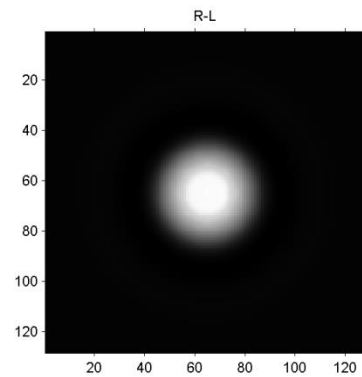
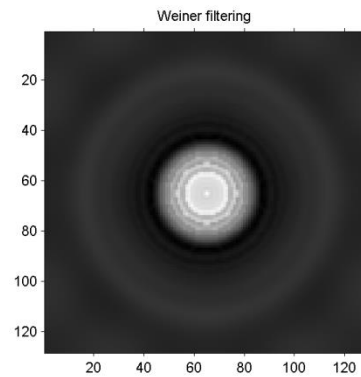
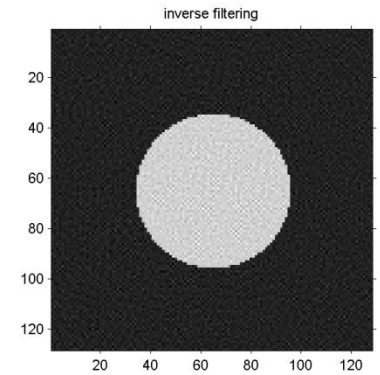
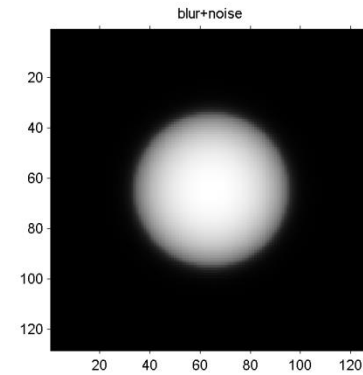
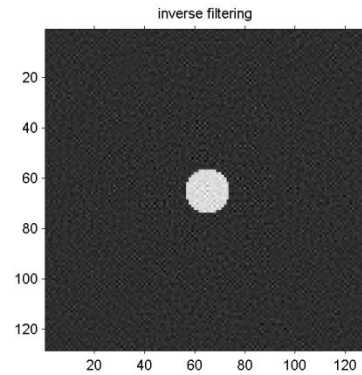
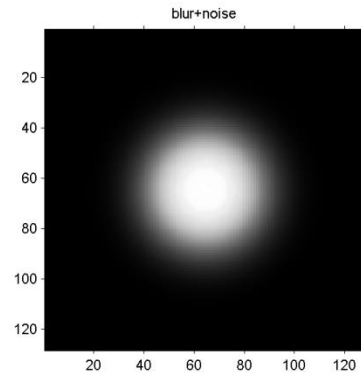
Examples



Examples



Examples



The end
