ECE 5984-Introduction to Modern Optical Microscopy

Homework-III

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In this report, the effect of change in spatial frequencies on the resolution of an image is analyzed. A square image with a circular dot in the middle is the object under consideration. The dot is 100nm in diameter and the square image has a dimension of 5 $\mu m \times 5$ μm. A step change of 10% to 90% is used as the parameter for calculating the resolution of the image.

Resolution calculation

To determine the resolution, the first step is to sum all the pixel values along x axis (or y axis) of the object. By this way, the integrated profile of the object along x-axis(or y axis) is obtained. Since, the object under consideration is circularly symmetric, the profile can be found along any direction.

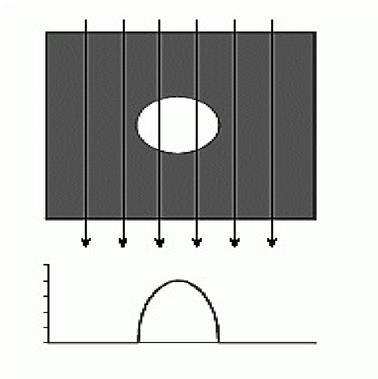

Figure 1: Integrated profile of the object (IP)

Once the integrated profile is obtained, the next step is to determine the step change by using the following equation,

$$Step(x) = \sum_{n=-\infty}^{x} IP(x)$$

The above equation is equivalent to integrating the profile of the object under consideration.

It is important to define "Resolution". In this report, resolution is defined as the minimum resolvable distance and is given by the 10% to 90% step distance. Hence, as we lose frequency content, we can expect the resolution(or the minimum resolvable distance) to increase.

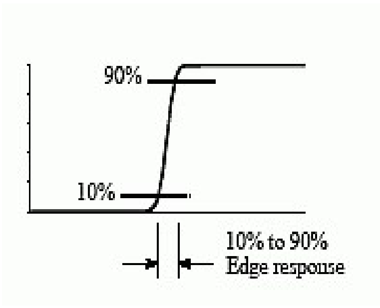

Figure 2: Step function of the object

Similar to one dimensional signals, low frequencies in images mean pixel values are changing slowly over space, while high frequency content means pixel values are rapidly changing in space. As we lose high frequency content of an image, it is expected for the pixel values in the image to change more gradually and thus the 10% to 90% distance will increase and so will the resolution(or the minimum resolvable distance).

SIMULATION RESULTS

In this section, the resolution is determined for an object with a circular aperture. The specifications used for the numerical simulation are as follows,

Radius of aperture, a	50 nm
Number of sample points, N	1024
Dimension of source plane	5 μm $\times$ 5 μm
Source plane grid spacing, δ_1	4.8829 nm
Illumination wavelength λ	600 nm

Figure 3: Simulation parameters

Part 1

In this part of the report, a circular aperture of radius 50 nm in a square image of dimension $5\mu m \times 5\mu m$ is taken as the input object. The Fourier transform of the object is computed to determine its frequency components and the resolution is determined using the procedure stated previously. The resolution will change depending on the number of sample points used. Because of the limitation on the computational power of my laptop, I have chosen 1024 samples.

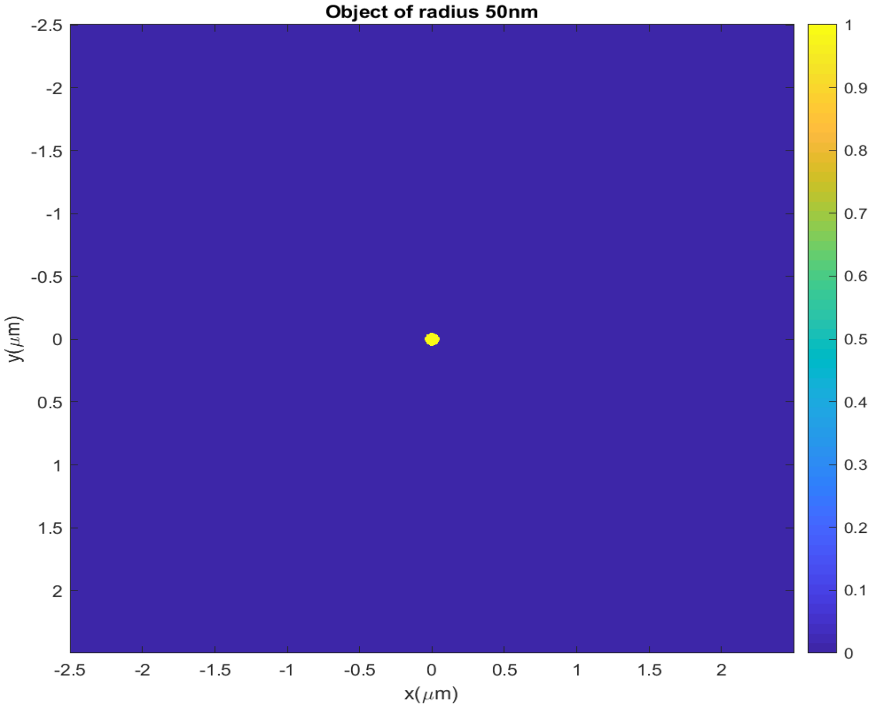

Figure 4: Object

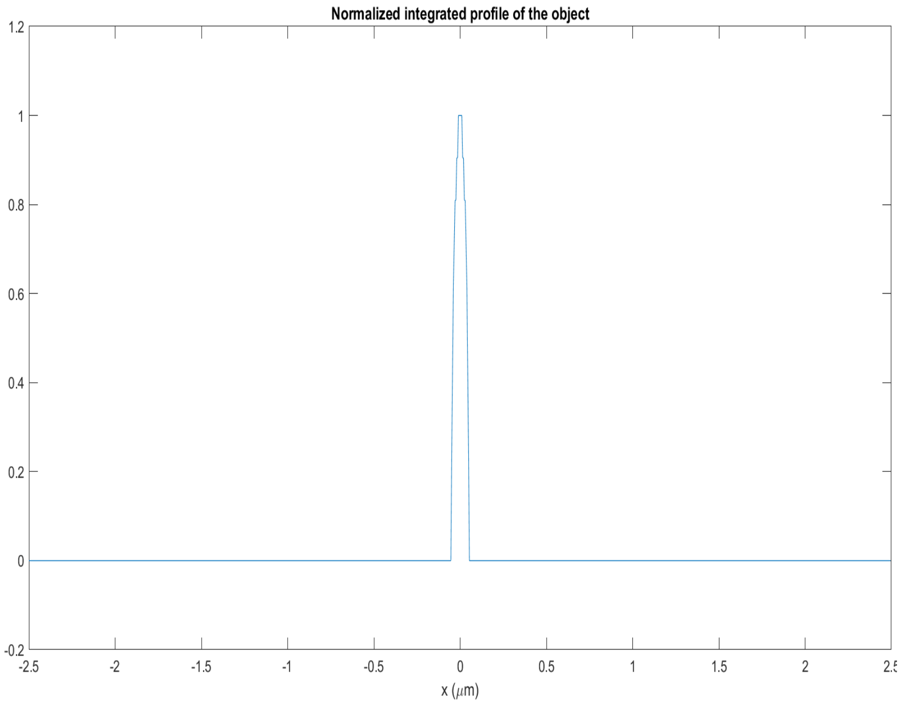

Figure 5: Integrated profile of the object

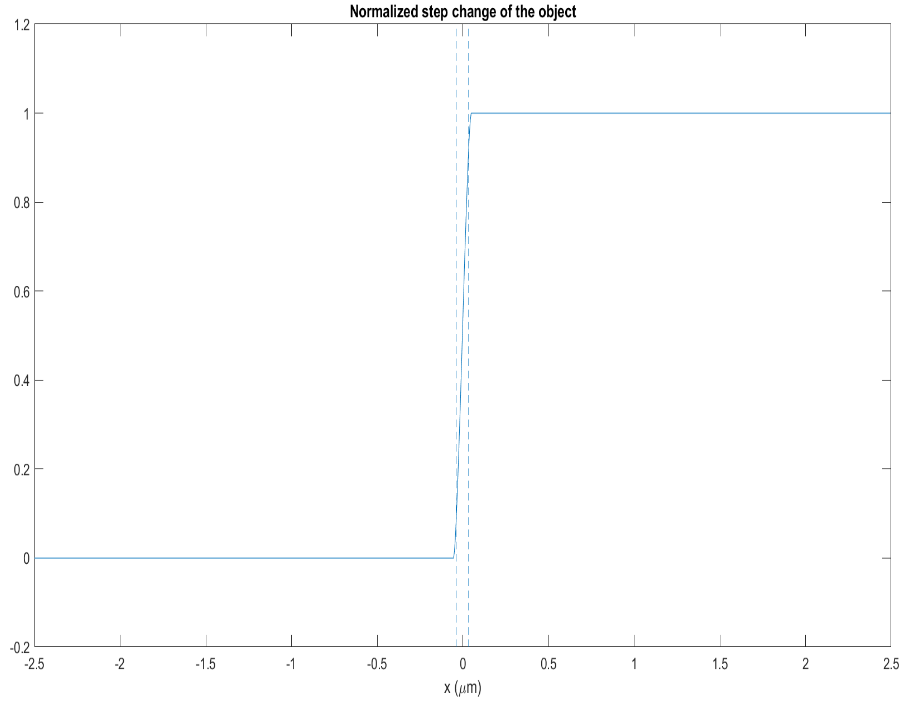

Figure 6: Step change of the object

According to the definition of resolution used in this report, the ideal resolution is zero.

The resolving power can be defined as the inverse of the distance it take to increase from 10% to 90% in the step change, then the ideal resolving power will be infinity (1/0).

Since the object under consideration is discretized, the resolution will not be zero. The resolution for the discretized object is **73.24219 nm** according to the step change in figure 6.

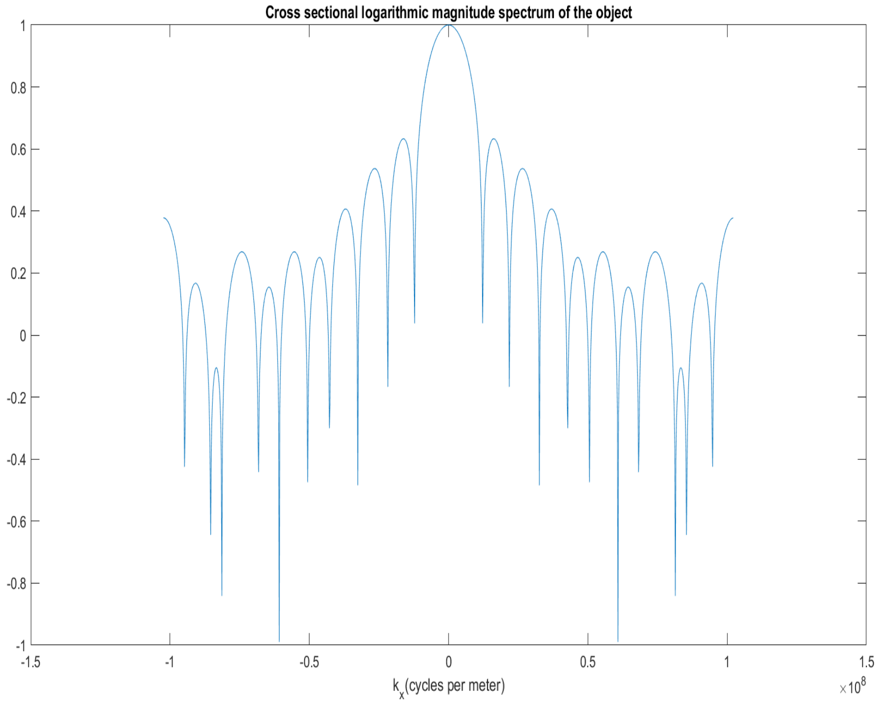

Figure 7: Normalized logarithmic magnitude spectrum of the object

In the magnitude spectrum of the object, the frequency spectrum can be visualized. It can be observed that the object has significant high frequency components. If the frequency spectrum of the object is modified, it can be expected to see a change in the object's spatial domain by carrying out an inverse Fourier transform .

Part 2

In this part, the amplitude field is propagated to far field and an image is formed. The image is analysed to determine its resolution and frequency content.

The object in the spatial domain can be represented as a spatial harmonic function in the x-y plane as follows,

$$f(x,y) = \iint F(v_x, v_y) exp[-j2\pi(v_x x + v_y y)]\, dv_x\, dv_y \tag{1}$$

with frequencies (v_x, v_y) and amplitudes $F(v_x, v_y)$. The spatial frequencies and the magnitude of the propagation vector in the x and y directions are related by $k_x = 2\pi v_x$ and $k_y = 2\pi v_y$.

When a plane wave of unity amplitude and wavelength(λ) traveling in the z direction is transmitted through the object with the circular aperture, the incoming unity plane wave is decomposed into many plane waves with different frequencies and corresponding propagation vectors. The transmitted wave is a superposition of plane waves,

$$U(x,y,z) = \iint F(v_x, v_y) exp[-j2\pi(v_x x + v_y y)] exp(-jk_z z)\, dv_x\, dv_y \tag{2}$$

Where, $k_z = \sqrt{k^2 - k_x^2 - k_y^2} = 2\pi\sqrt{\lambda^{-2} - v_x^2 - v_y^2}$. The magnitude of the propagation vector is $k = 2\pi/\lambda$.

For k_z to be real, v_x and v_y must be less than $1/\lambda$. Frequencies greater than this value are evanescent and will not propagate to the image plane at far field. Thus, we can expect to lose frequency information above $1/\lambda$ at the image plane at far field. The minimum resolvable distance will be greater than the one in the object plane because of this. The required cut-off window is created to filter the frequencies above the cut-off and then the image is formed by performing an inverse Fourier transform.

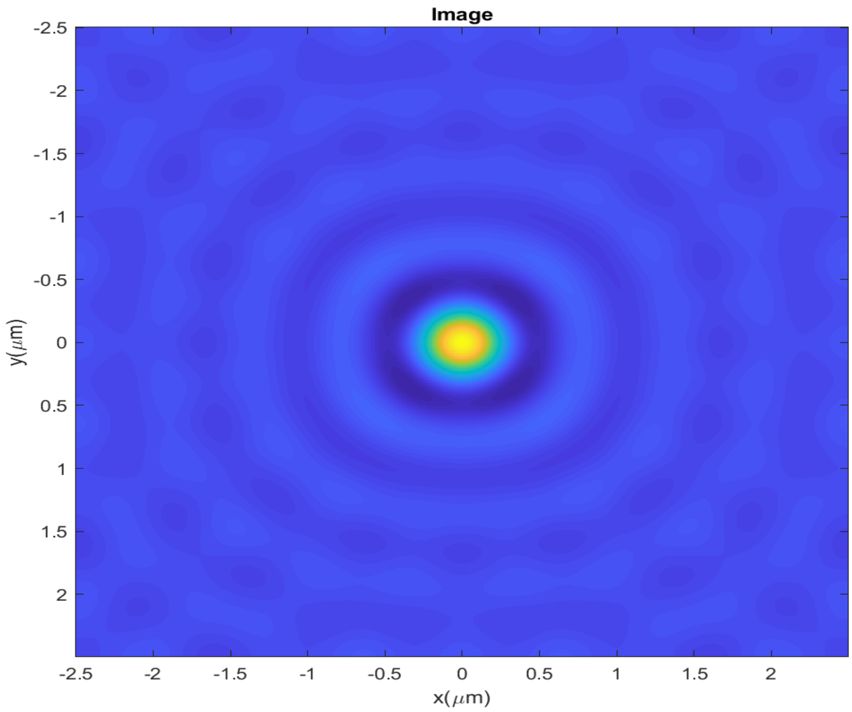

Figure 8: Image at far field

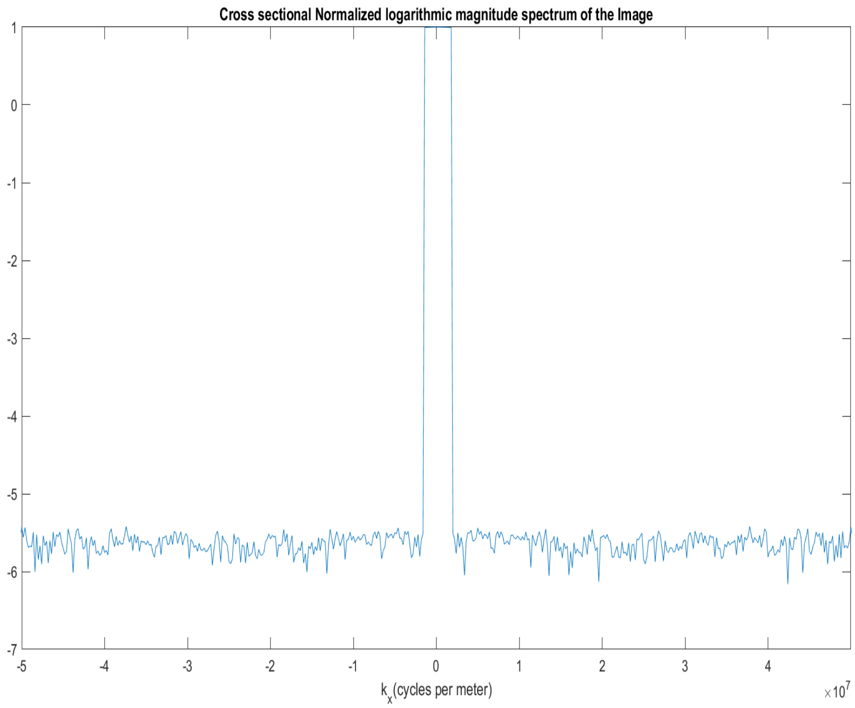

Figure 9: Frequency spectrum of the Image

The cut-off frequency is $1/\lambda = 1/(600 \times 10^{-9}) = 0.167 \times 10^7$. As expected, it can be observed that in the frequency spectrum, frequencies above 0.167×10^7 have been cut off at the far field image plane. Because of the loss of high frequency components, it can be seen that image is not similar to the one at the object plane. The sharp structure and the boundaries in the spatial domain are not present because of the absence of the high frequency components.

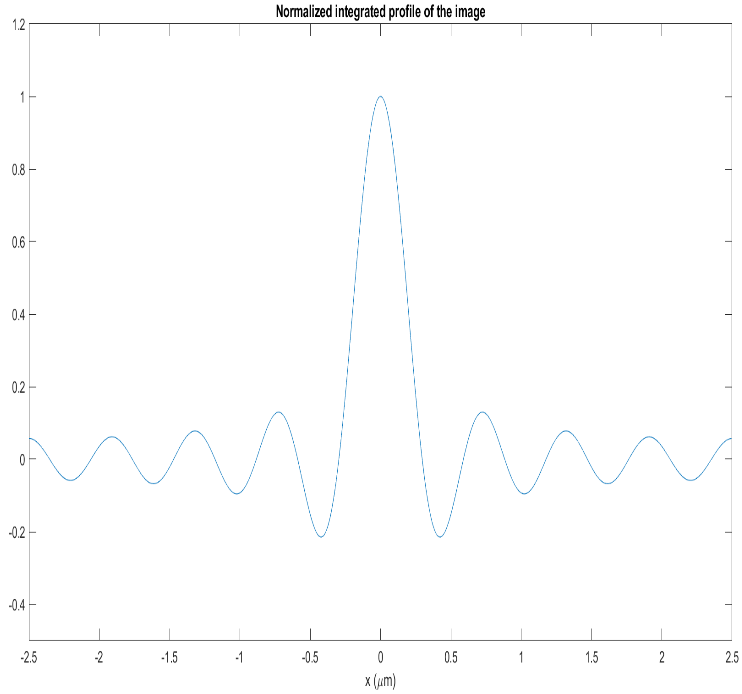

Figure 10: Integrated profile of the Image

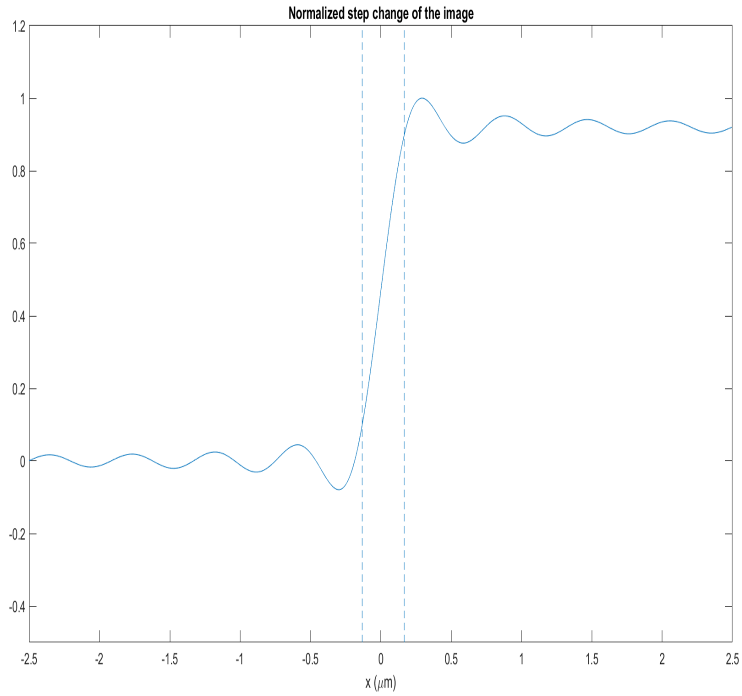

Figure 11: Step change of the Image

The resolution in this case is **0.29785** μ**m** which is greater than the one obtained in the previous case. This means that two points have to be separated by a minimum of 297.85 nm to be resolved. On the other hand, in the previous case, it was only 73.2419 nm. The increase in resolution(or the minimum resolvable distance) is because of the loss of high frequency components.

Part 3

In the previous part, it was shown that the object when illuminated by an incident plane wave, the transmitted wave is a superposition of plane waves travelling with different propagation vectors. The maximum spatial frequency that will propagate is $1/\lambda$. This is a direct consequence of the limit provided by the equation $k_z = \sqrt{k^2 - (k_x^2 + k_y^2)}$. For k_z to be real, v_x and v_y should be less than $1/\lambda$. Frequencies greater than this value are evanescent and will not propagate to the image plane at far field. In addition to the above limit, here it is provided that the field is collected by a lens within an angular range +/- 30 degree around the optical axis, and then the image is formed. The transmitted wave has plane waves with frequencies (v_x, v_y) that travel at angles, with respect to the optic axis, given by

$$\theta_x = sin^{-1}(\lambda v_x) \tag{3}$$

$$\theta_y = sin^{-1}(\lambda v_y) \tag{4}$$

Thus, we have

$$v_x = sin(\theta_x)/\lambda \tag{5}$$

$$v_y = sin(\theta_x)/\lambda \tag{6}$$

For an angle of θ= 30 degrees around the optic axis, the maximum spatial frequency collected by the lens will be $v_{x_{max}} = v_{y_{max}} = 1/(2\lambda) = 0.8333 \times 10^6$. Thus, we can expect to lose frequency information greater than $1/(2\lambda)$ at the far field image plane. The resolution(or the minimum resolvable distance) will be even greater than the previous case because of the frequency information loss.

The required cut-off window is created to filter the frequencies above the cut-off and the image is formed.

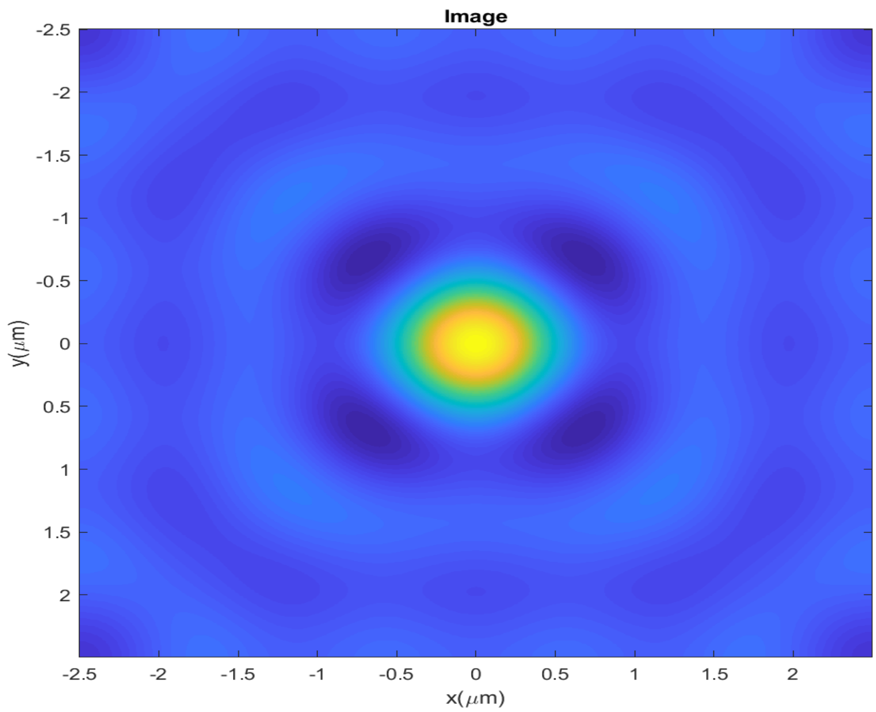

Figure 12: Image at far field

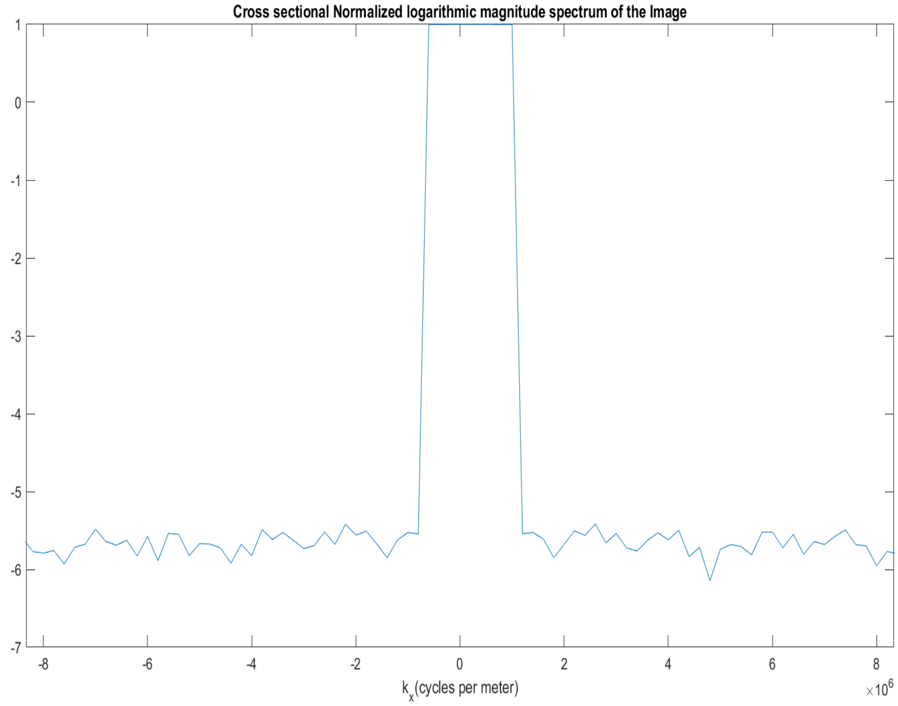

Figure 13: Frequency spectrum of the Image

The cut-off frequency is $1/(2\lambda) = 1/(2*600\times10^-9) = 0.833\times10^6$. As expected, it can be observed that in the frequency spectrum, frequencies above 0.833×10^6 have been cut-off.

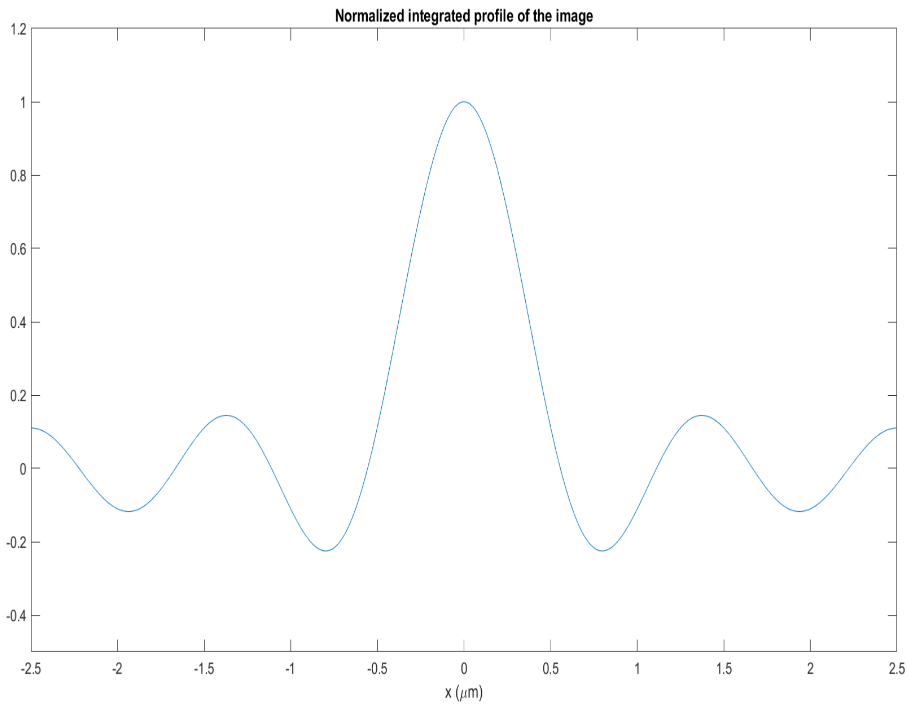

Figure 14: Integrated profile of the Image

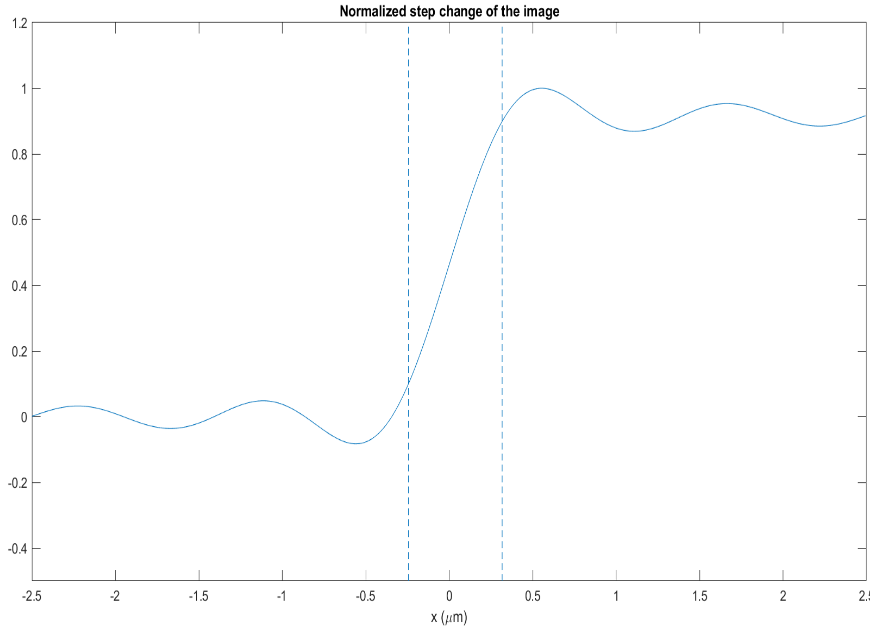

Figure 15: Step function of the Image

The resolution in this case is **0.561523 μm or 561.523 nm** which is greater than the ones obtained in previous cases. As we lose frequency information, an increase in the minimum resolvable distance is observed starting from 73.4219 nm in the first case, 297.85nm in the second case to 561.523 nm in the third case.

Part 4

In this part, a modulation transfer function is added to the frequency distribution of the image obtained in the previous section. The MTF is such that the transmission is linearly reduced along the radial direction. To implement this condition, a radially symmetric triangular function is used.

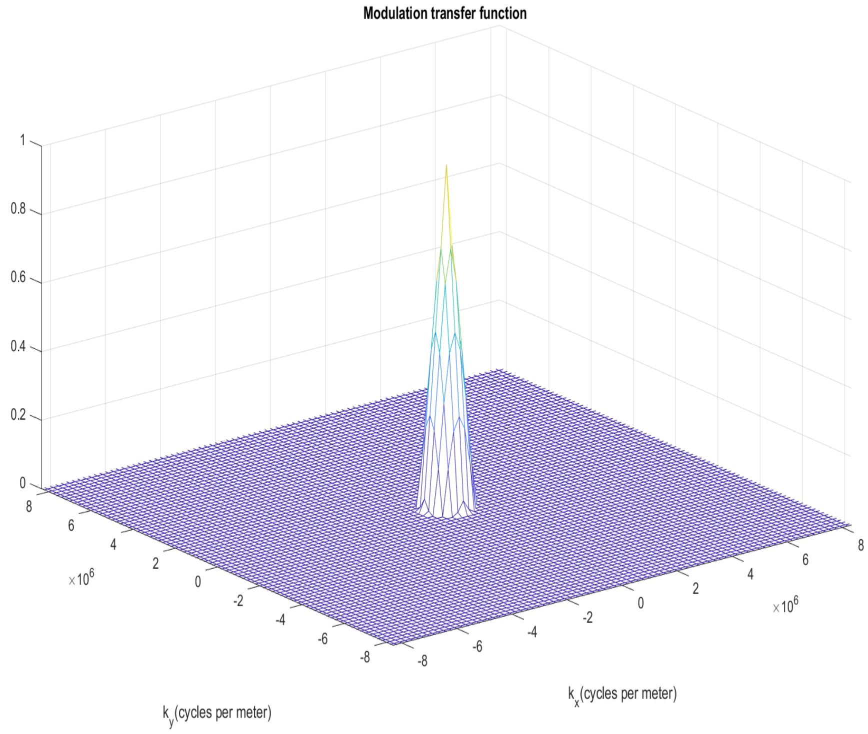

Figure 16: 3D Modulation Transfer Function

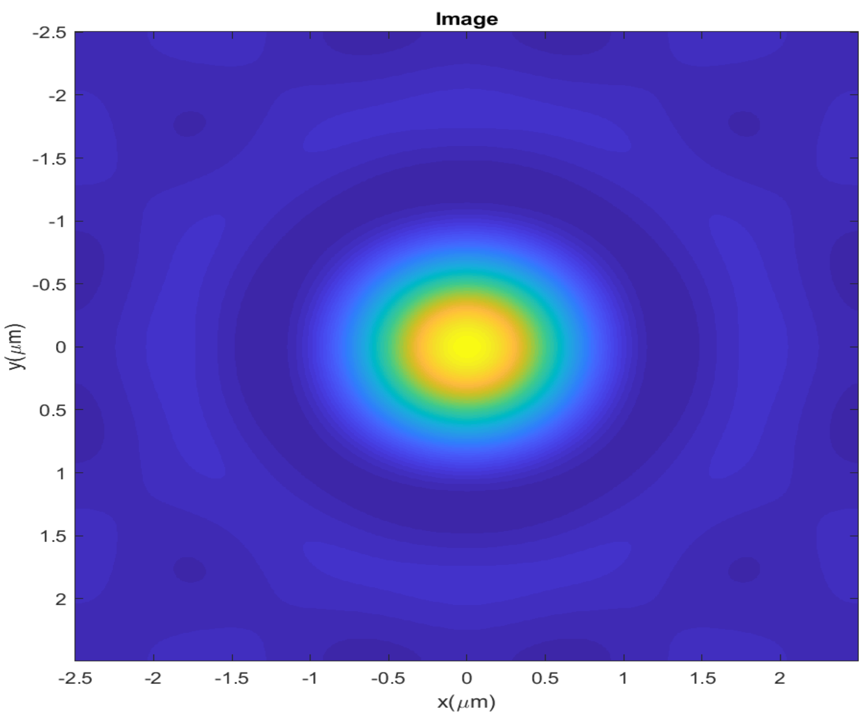

Figure 17: Image at far field

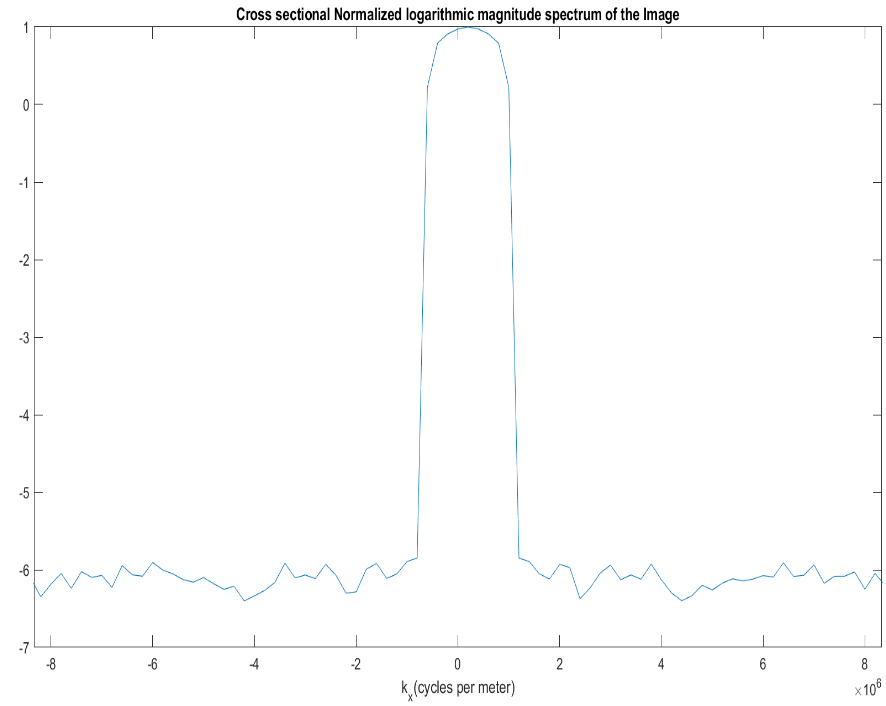

Figure 18: Frequency spectrum of the Image

The cut-off frequency is $1/(2\lambda) = 1/(2*600\times 10^-9) = 0.833\times 10^6$. As expected, it can be observed that in the frequency spectrum, frequencies above 0.833×10^6 have been cut-off. Unlike the previous case, here the pass band frequencies are linearly attenuated by the radially symmetric triangular function. In the previous case, pass-band frequencies have uniform amplitude.

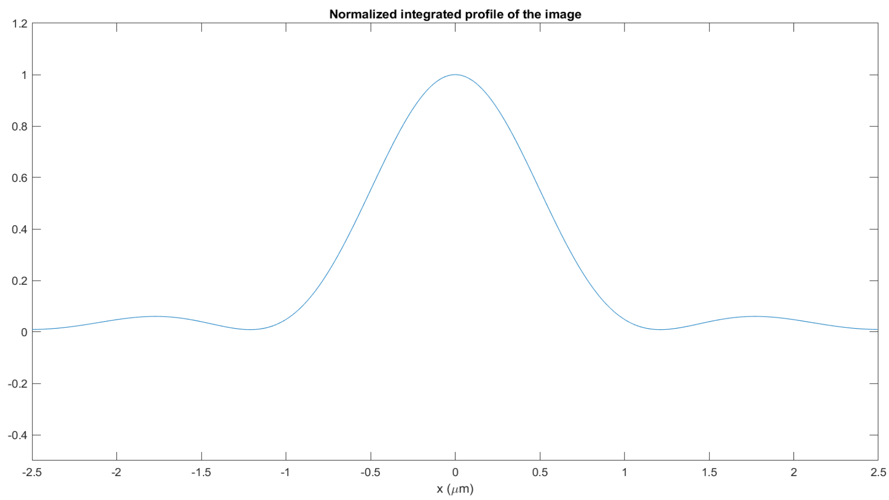

Figure 19: Integrated profile of the Image

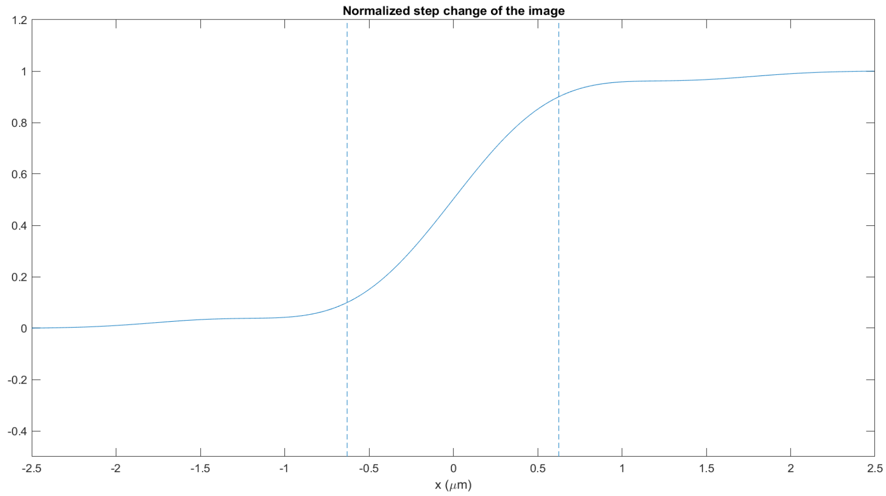

Figure 20: Step change of the Image

The resolution is **1.25488 μm** or **1254.88 nm** which is greater than the previous cases. This is expected because the attenuation of frequencies in the pass band will result in frequency information loss and the pixel variation will be even more gradual. Thus, the step change will widen and the minimum resolvable distance or resolution will increase.

Part 5

In this part, a deconvolution is carried out on the image obtained using the modulation transfer function in section 4 to recover the image obtained in section 3.

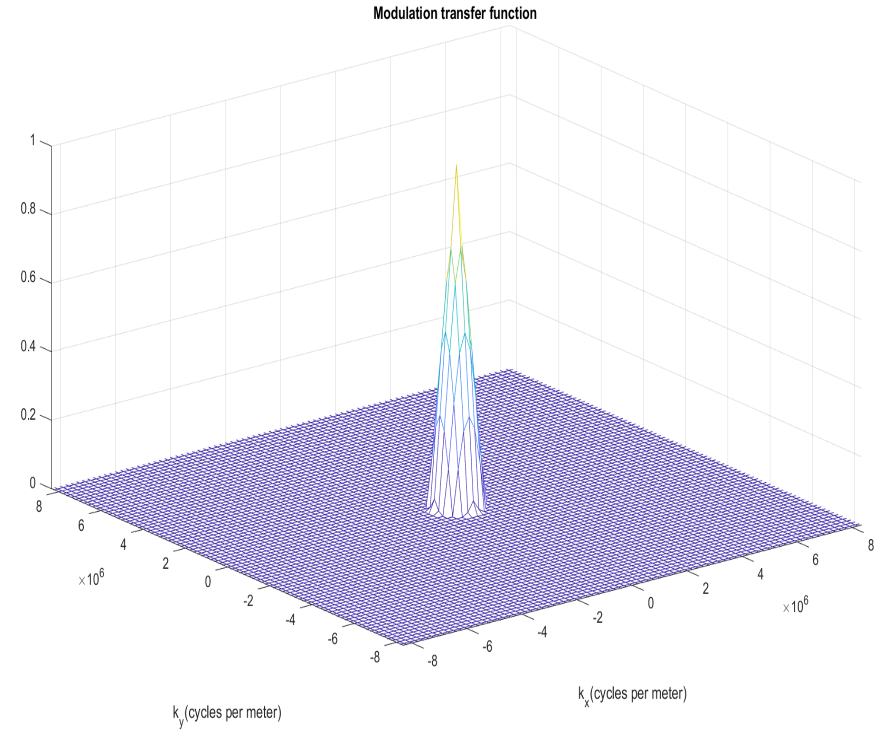

Figure 21: MTF

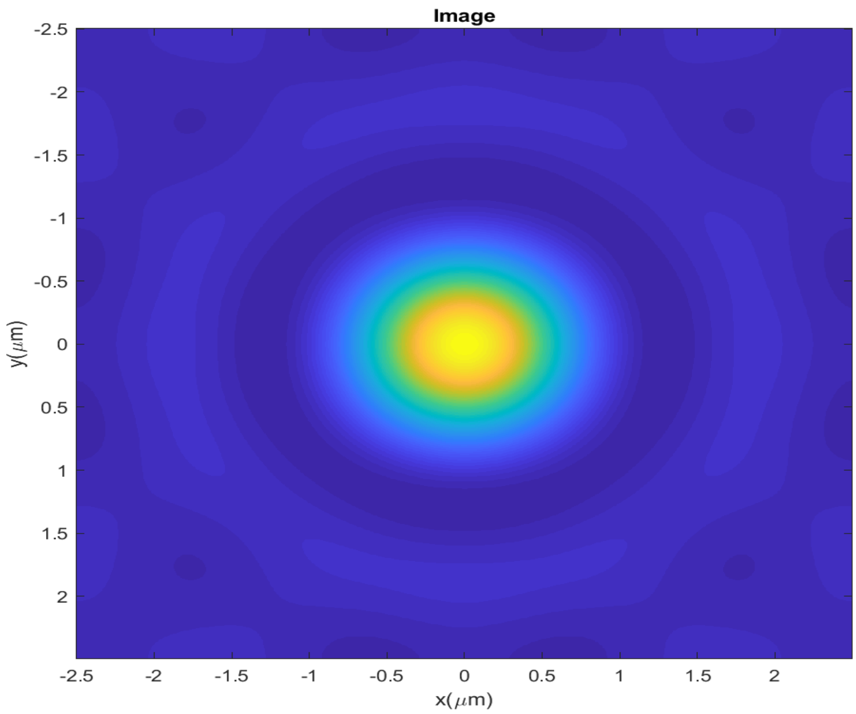

Figure 22: Image at far field

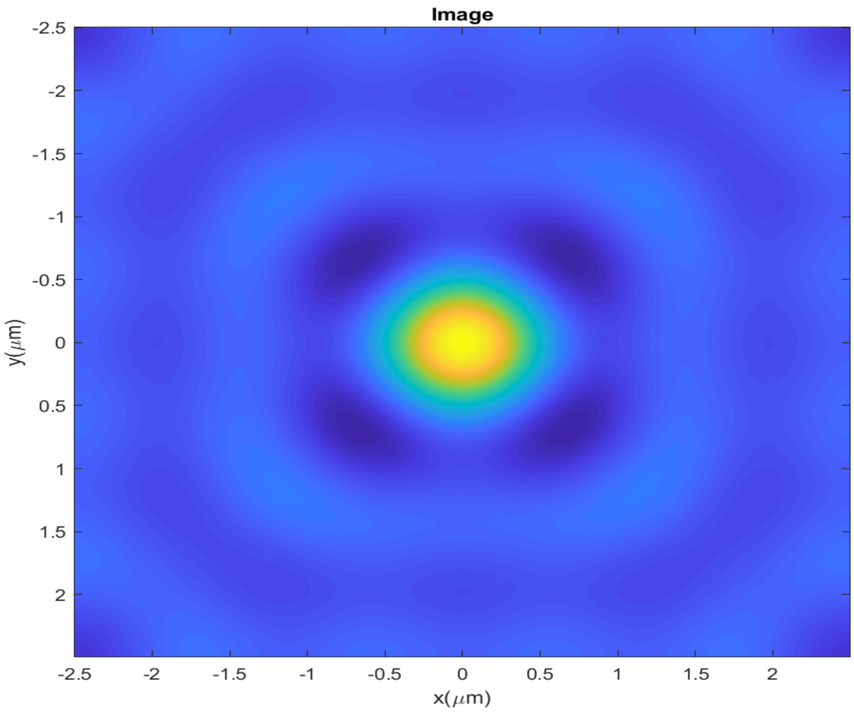

Figure 23: Deconvolved image

The deconvolved image is identical into to the image obtained in figure 12 in part 3. The deconvolution is carried out in the absence of noise, hence we are able to recover the exact image. With added noise, the spatial frequencies in the input image will change and the deconvolved image will not be identical to the original image.

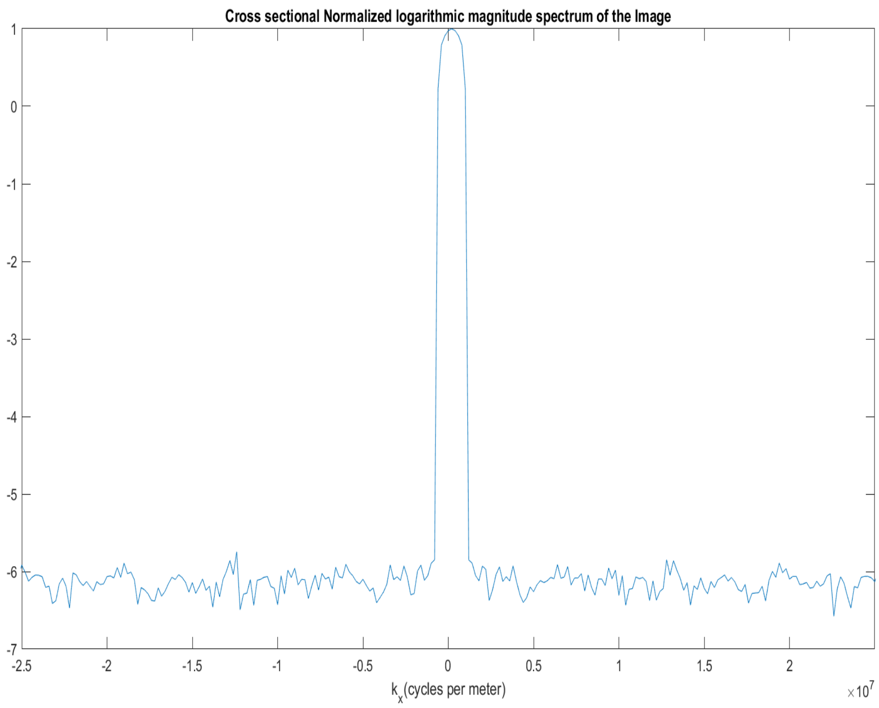

Figure 24: Frequency spectrum of the Image obtained using MTF

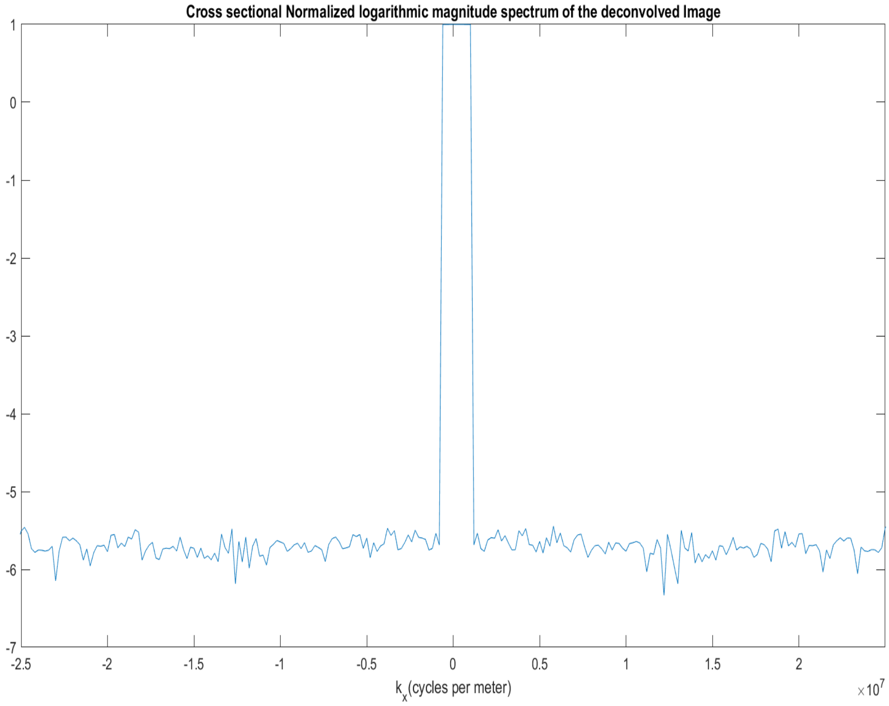

Figure 25: Frequency spectrum of the deconvolved Image

The resolution of the image obtained with the modulation transfer function is **1254.88 nm** and the resolution of the image obtained using deconvolution is **561.5234 nm**. Deconvolution provides an improvement in resolution

Part 6

In this part, the deconvolution is carried out on the image but this time white Gaussian noise is added to the image before deconvolution. Varying levels of noise are added to the input image(image obtained in part 3) before deconvolution is carried out to analyse how the noise impacts the deconvolution process.

The noise is a white Gaussian noise with mean $\mu = 0$ and variance corresponding to a specific Signal-to-Noise (SNR). Initially, the mean square value of the input image(image obtained in part 3) X is computed as follows,

$$\text{Mean square value} = \sum X^2$$

With the computed mean square value of the image, the variance of the image is computed as follows,

$$\sigma^2 = \frac{\Sigma X^2}{N} - \mu^2$$

Where, 'N' is the total number of elements in the 2D matrix and μ is the mean value. Since $\mu = 0$, the variance of the image is given by,

$$\sigma^2 = \frac{\Sigma X^2}{N}$$

The variance of the Gaussian noise is determined as follows,

$$SNR_{dB} = 10 \times log\left[\frac{variance(image)}{variance(noise)}\right]$$

For a given image and SNR, the variance of the noise is:

$$Var(noise) = \frac{Var(image)}{10^{\frac{SNR}{10}}}$$

It is of importance that the original image should be normalized by dividing it by the maximum pixel value before variance computation. If not, the variance of the noise will not be a value in the range from 0 to 1. The white Gaussian noise is added to the image using 'Imnoise' function.

For instance, if SNR=10dB, the variance of the noise is $\frac{Var(image)}{10}$

If SNR=50dB, the variance of the noise is $\frac{Var(image)}{10^5}$

Thus, varying levels of noise can be added to the image and an analysis can be carried out to determine the effect on the deconvolution output.

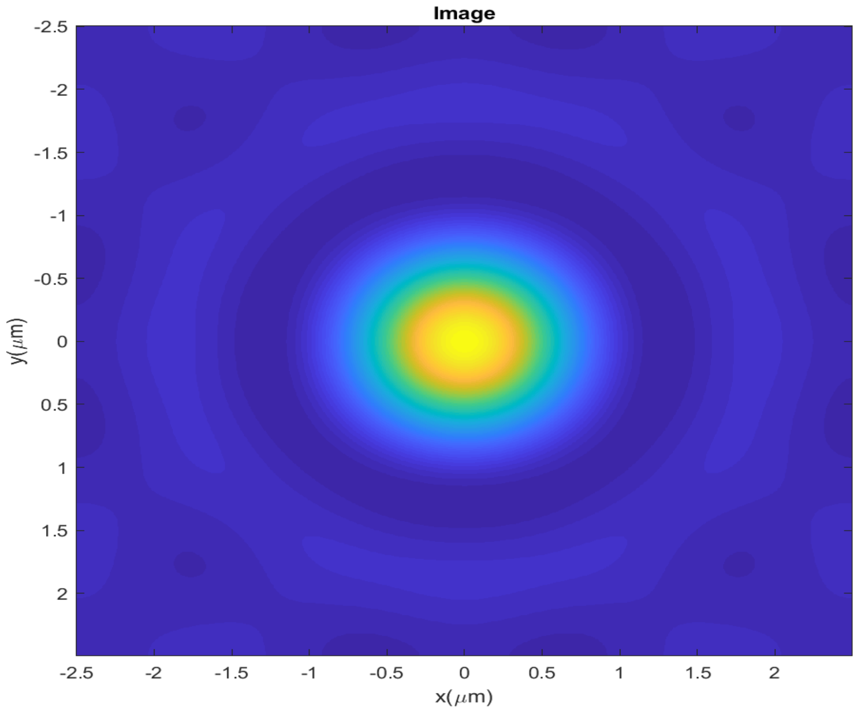

Figure 26: Noise free input image for deconvolution process

Resolution before deconvolution - 1.254883 μm

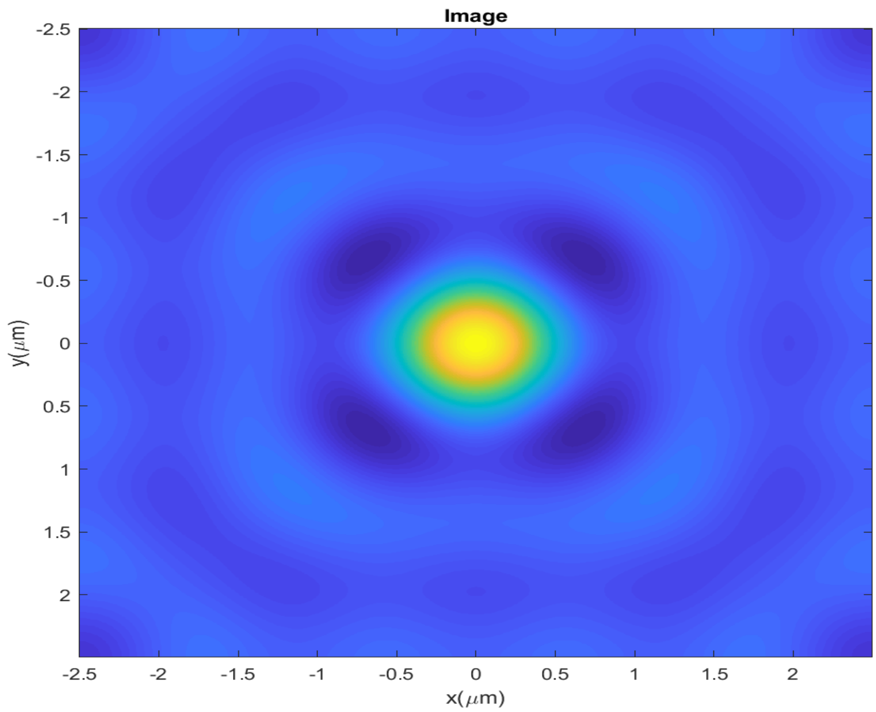

Figure 27: Deconvolved image

Resolution after deconvolution - 0.5615234 μm

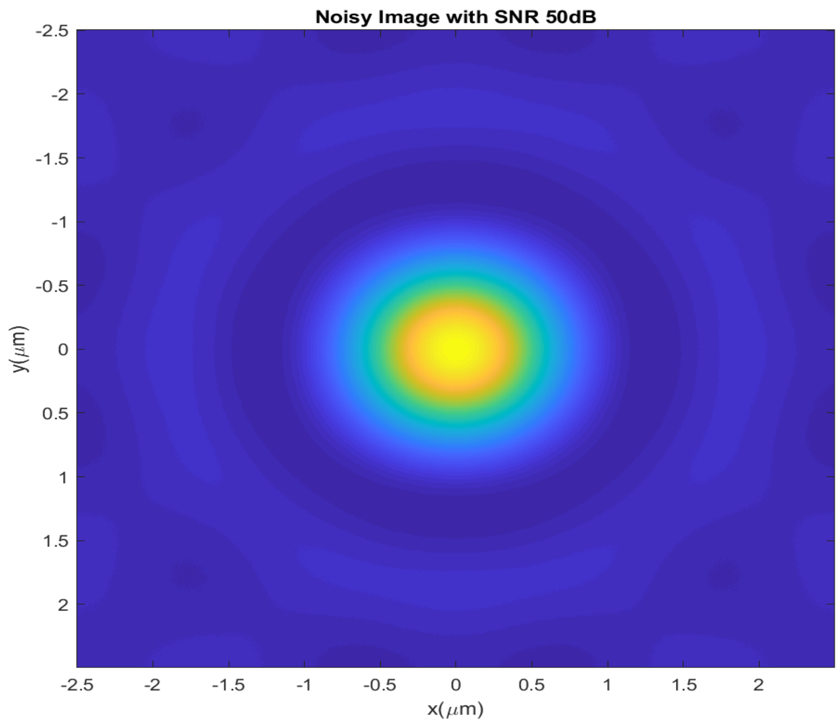

Figure 28: Input image for deconvolution with SNR=50dB added noise

Resolution before deconvolution - 2.480469 μm

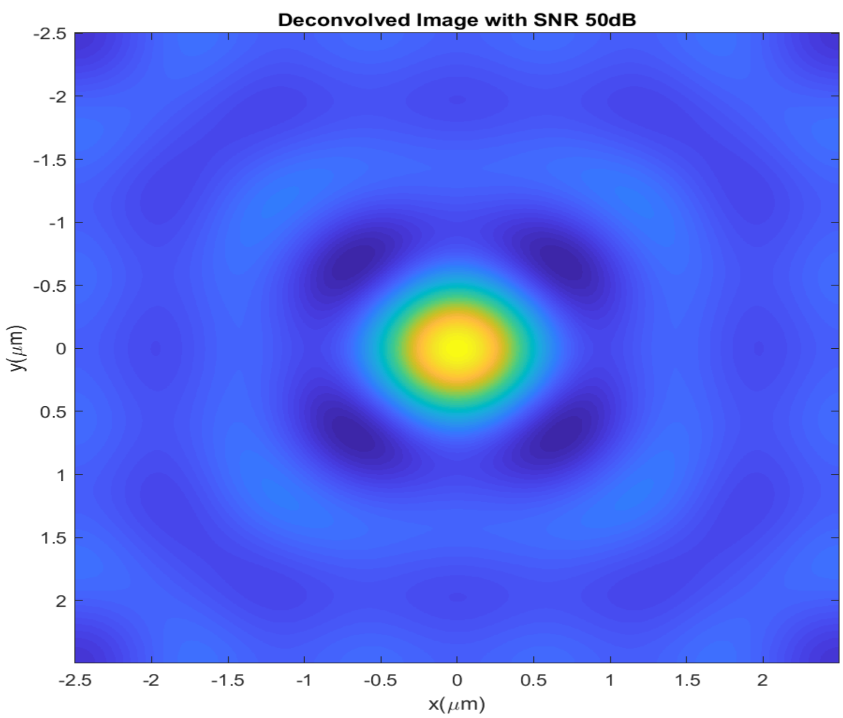

Figure 29: Deconvolved image with SNR=50dB

Resolution after deconvolution - 1.625977 μm

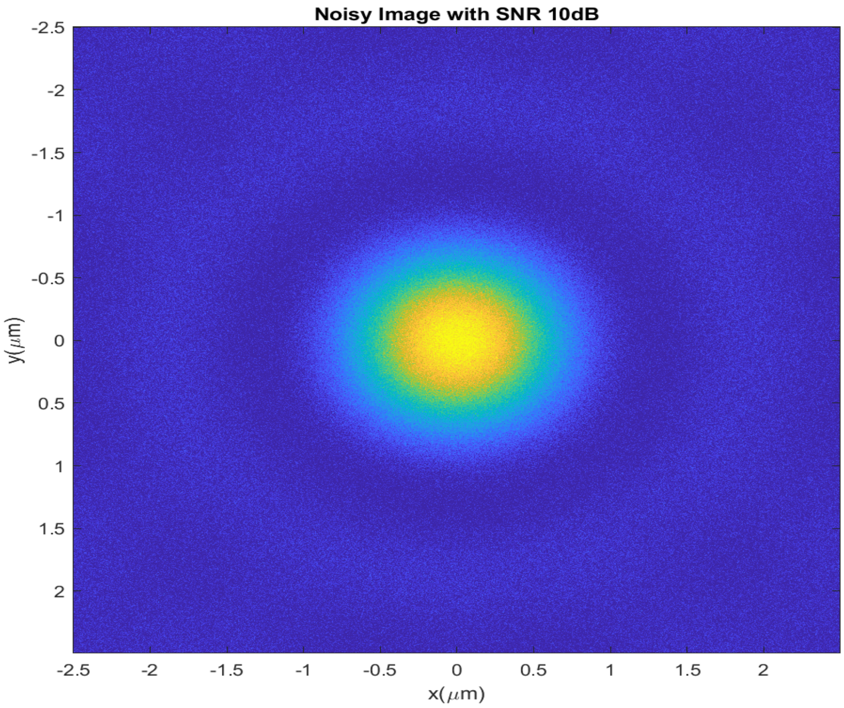

Figure 30: Input image for deconvolution with SNR=10dB added noise

Resolution before deconvolution - 2.856445 μm

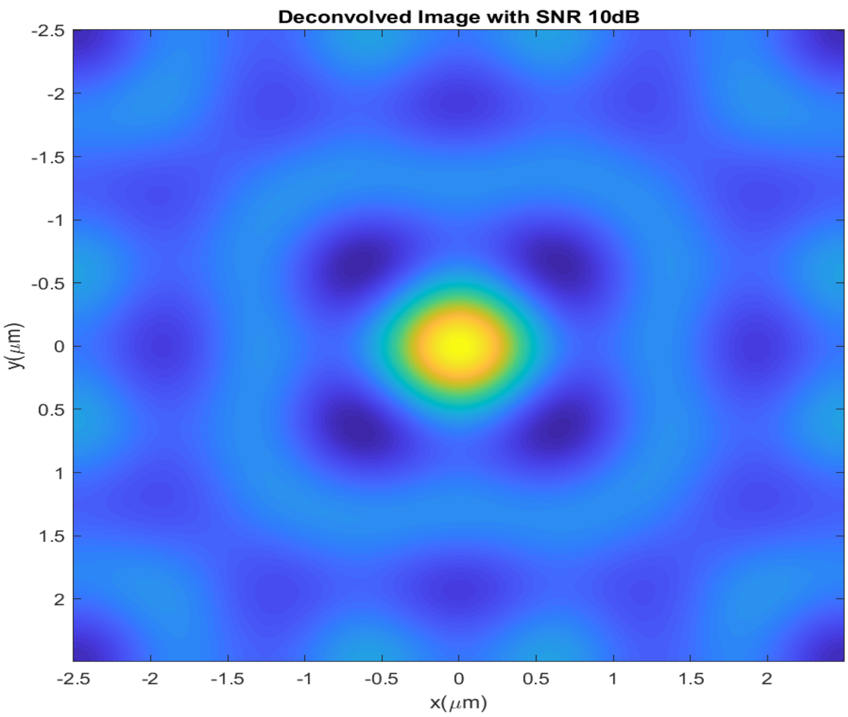

Figure 31: Deconvolved image with SNR=10dB

Resolution after deconvolution - 2.016602 μm

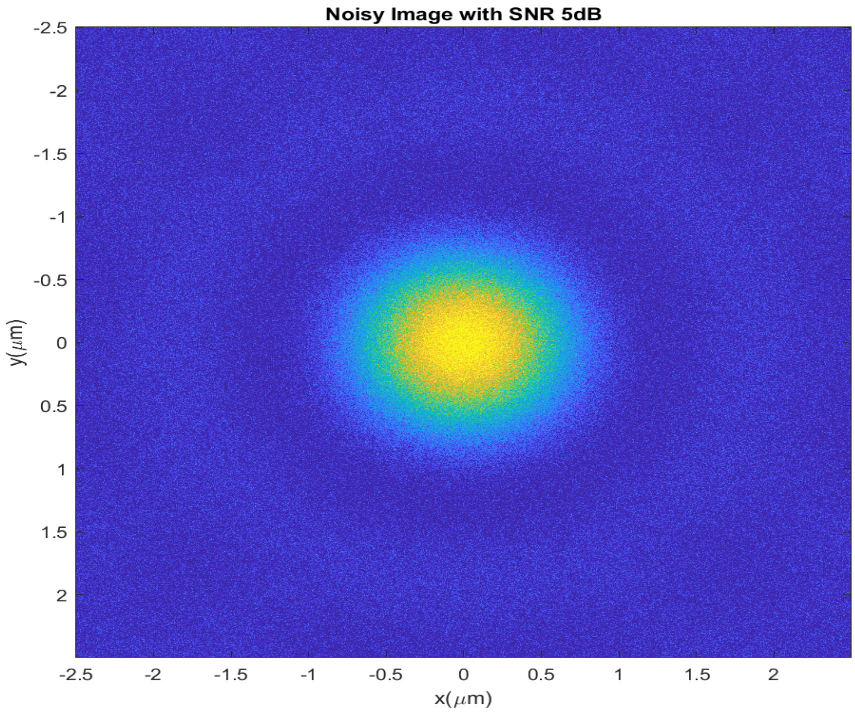

Figure 32: Input image for deconvolution with SNR=5dB added noise

Resolution before deconvolution - 3.178711 μm

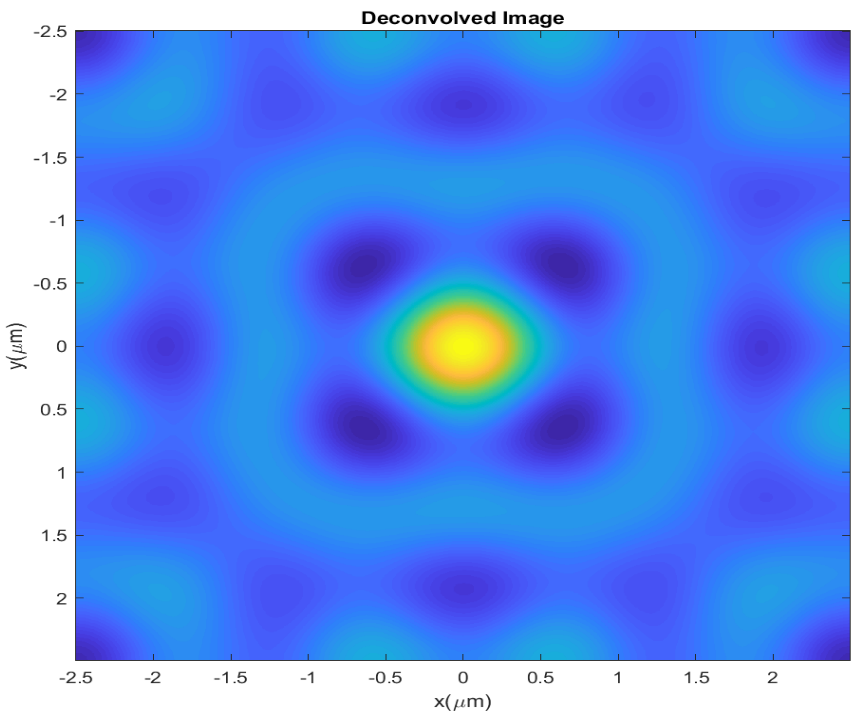

Figure 33: Deconvolved image with SNR=5dB

Resolution after deconvolution - 2.734375 μm

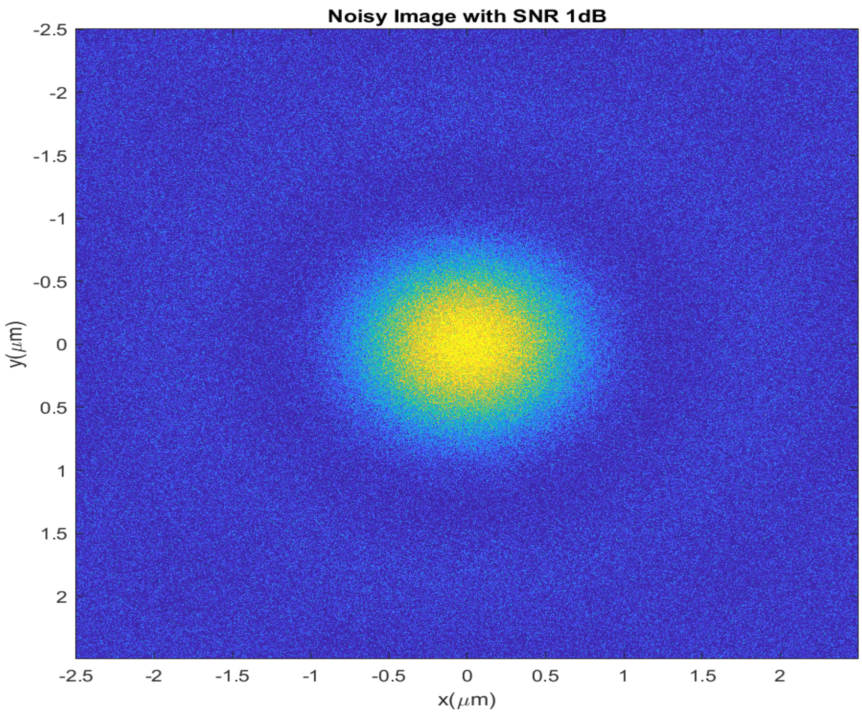

Figure 34: Input image for deconvolution with SNR=1dB added noise

Resolution before deconvolution - 3.422852 μm

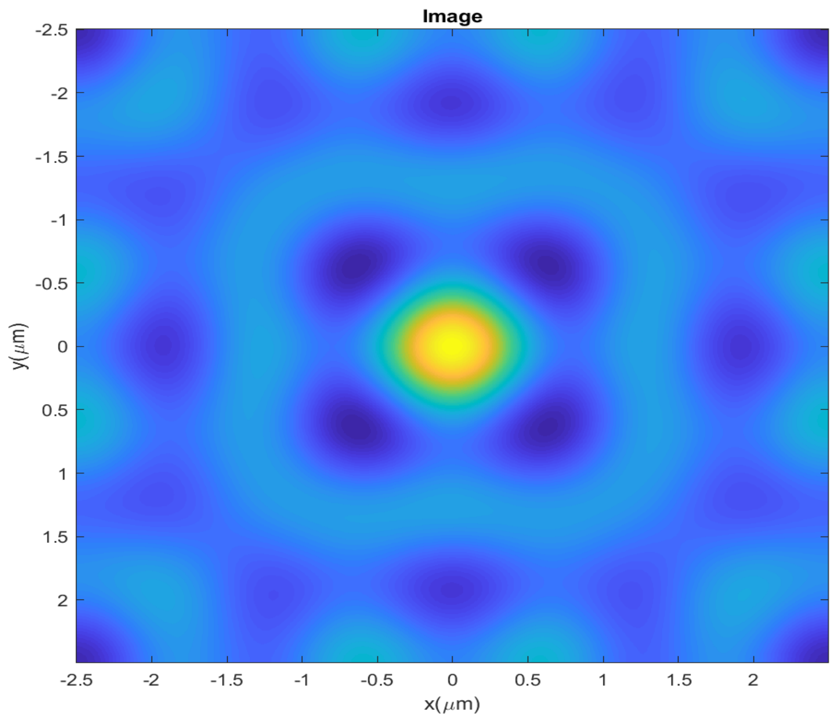

Figure 35: Deconvolved image with SNR=1dB

Resolution after deconvolution - 2.900391 μm

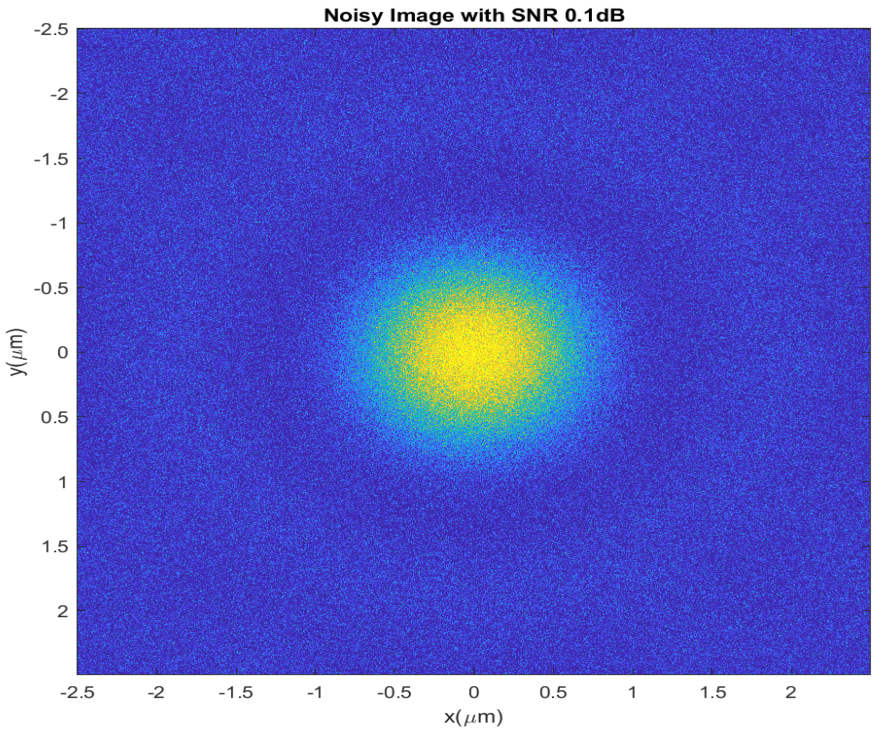

Figure 36: Input image for deconvolution with SNR=0.1dB added noise

Resolution before deconvolution - 3.471680 μm

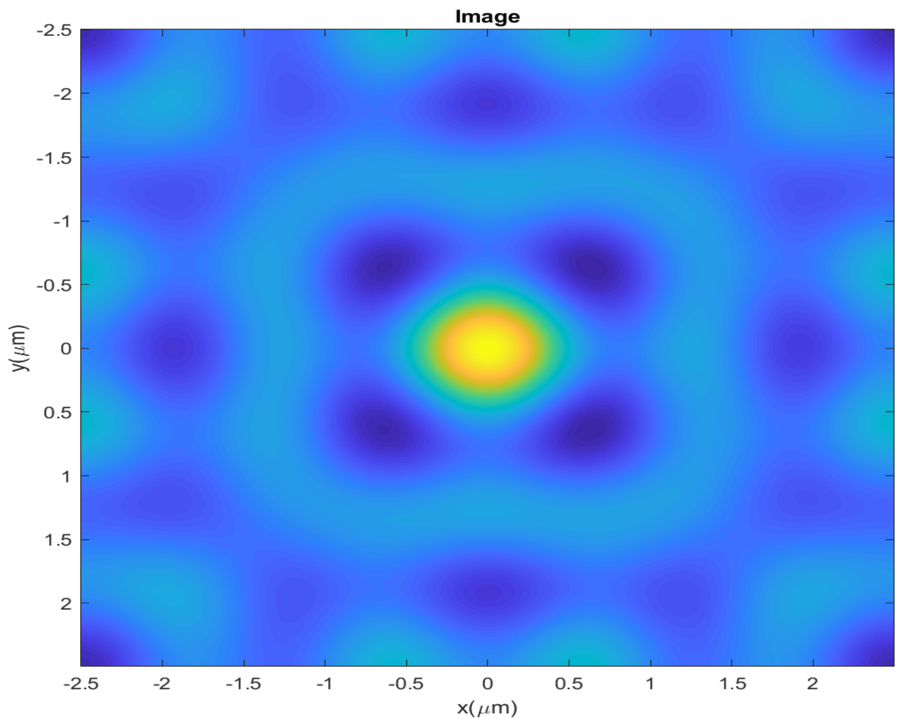

Figure 37: Deconvolved image with SNR=0.1dB

Resolution after deconvolution - 2.958984 μm

It is observed that as the Signal to noise ratio is decreased, the spatial domain signal is significantly corrupted by noise. This means the frequency components will be altered too. Thus, when the deconvolution is carried out on this noisy image, the recovered output image is not identical to the deconvolved image obtained from a noise free image. Also, it is found the resolution or minimum resolvable distance increases as noise power increases. This is expected because as the noise power increases, the frequency spectrum within the pass band will be altered significantly. For instance, the frequency spectrum of the deconvolved image with added noise such that SNR=0.1dB is as follows,

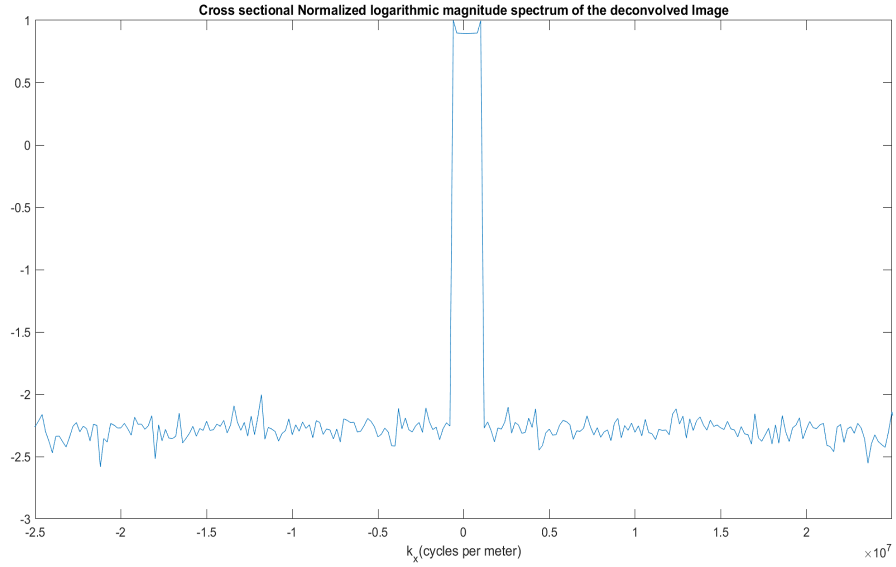

Figure 38: Deconvolved image spectrum with SNR=0.1dB

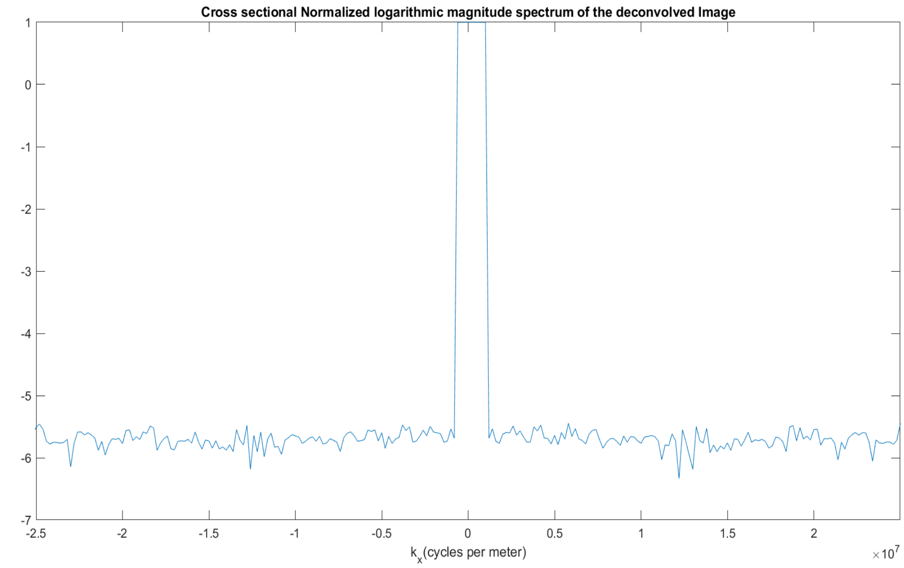

Figure 39: Deconvolved image spectrum with zero noise

By comparing the above two images, it is observed that the deconvolved image with added noise has its frequency spectrum modified in the passband. Thus, it is concluded that the added noise modifies the frequency spectrum of the image and thus the deconvolved image with added noise will not be identical to the expected image.