Background Information:

The background information provided here and basic 2D signals and systems knowledge is all you need to work on the tasks described in the section that follows. Try to apply your knowledge to derive your own answers and solutions and resist the temptation to copy information available in the literature or through google search. Nevertheless, you are encouraged to gather information that will help you do your best on the tasks described below. Be sure to list all references you have consulted.

A two-dimensional (2D) image, which is formed by a conventional widefield (WF) microscope, can be modelled as the 2D convolution of the underlying object, $o(x, y)$, and the Point Spread Function (PSF), $h(x, y)$, of the microscope (which is the impulse response of the imaging system) as follows:

$$g_{WF}(x, y) = o(x, y) \otimes_2 h(x, y) \tag{1}$$

where (x, y) denotes the lateral coordinates in the space domain, and $\otimes_2$ denotes the 2D convolution operation. Eq. (1) is called the 2D WF forward imaging model or 2D forward WF

image. Note that the 2D spatial-frequency Fourier transform of the PSF, known as the optical transfer function (OTF), is a bandlimited filter that characterizes the imaging system. Object information is transferred by the imaging system only within the band pass of this OTF filter. All other information is not transferred and thus not imaged.

The 2D image formed by a 2D structured illumination microscope (2D-SIM), can be modelled as a 2D convolution of the product of the object and the illumination pattern, $o(x, y) i(x, y)$, and the PSF, $h(x, y)$, as follows:

$$g_{SIM}(x, y) = \left[o(x, y) i(x, y) \right] \otimes_2 h(x, y) \tag{2}$$

where the illumination pattern in 2D-SIM is a sinusoidal pattern as follows:

$$i(x, y) = 1 + \cos \left[2\pi u_m \left(x \cos \theta + y \sin \theta \right) + \varphi \right] \tag{3}$$

and φ is the phase of the pattern and θ defines the orientation of the pattern with respect to the x axis. Eq. (2) is called the 2D SIM forward imaging model or 2D forward SIM image.

Tasks:

1. Please provide a mathematical derivation that shows what is the relation between the images $g_{WF}(x,y)$ and $g_{SIM}(x,y)$ defined by Eqs. (1) and (2) above. For this task, you can assume, without loss of generality, that $\varphi = 0$, $\theta = 0$ for simplicity. **Hint:** Consider investigating the Fourier content of the images formed by the SIM and WF microscopes.
2. Based on your derivation in Task 1, please show and/or describe using equations and figures what you believe might be the additional information gained with SIM imaging from the use of the sinusoidal illumination versus the use of uniform illumination used in WF imaging. In this part you should try to explain why SIM imaging is better in some sense than WF imaging. Provide as much detail as possible to explain your understanding.
3. A link was provided in the email for a .mat file that contains three 2D forward images from a 2D-SIM system, which have a different phase, φ defined in Eq. (3), i.e.:

$$g_{SIM,1}(x,y) = \left\{ o(x,y)\left[1+\cos\left(2\pi u_m x + \varphi_1\right)\right]\right\} \otimes_2 h(x,y)$$

$$g_{SIM,2}(x,y) = \left\{ o(x,y)\left[1+\cos\left(2\pi u_m x + \varphi_2\right)\right]\right\} \otimes_2 h(x,y)$$

$$g_{SIM,3}(x,y) = \left\{ o(x,y)\left[1+\cos\left(2\pi u_m x + \varphi_3\right)\right]\right\} \otimes_2 h(x,y)$$

in which $\varphi_1 = 0°$, $\varphi_2 = 120°$, $\varphi_3 = 240°$, respectively. In addition, a 3×3 matrix is given which is formulated as follows:

$$\mathbf{M} = \begin{bmatrix} 1 & 0.5e^{i\varphi_1} & 0.5e^{-i\varphi_1} \\ 1 & 0.5e^{i\varphi_2} & 0.5e^{-i\varphi_2} \\ 1 & 0.5e^{i\varphi_3} & 0.5e^{-i\varphi_3} \end{bmatrix}$$

Please write a Matlab script to decouple any new information captured with a SIM system, which is not present in the WF image, from the WF image. Your program should take as input the 3 2D-SIM images and provide as output the WF image and any other information present in the SIM images. Provide a copy of your Matlab script and figures of the input and output images in both the space and spatial-frequency domains. All figures should appear in a report with appropriate figure captions and a discussion explaining them. Your code should be ready and easy for me to run so that I can verify that it works. Be sure to label all figures with appropriate titles and to display scale bars that show the intensity values in images.