

# Tutorial 1: First *in-silico* microscopy image

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6 Jul, 2021

## 1 Generate the the PSF

The point spread function  $PSF(l', m', n')$  is generted using the following command

```
Tut1$ siliscopy gen_psf --method gandy --paramfile parameters.  
dat --calc all --output PSF_gandy --multiprocess
```

This reads the following variables from `parameters.dat`,

- `NA` = 1.3 (Numerical Aperture)
- `meu` = 1.51 (Refractive index of immersion oil)
- `d1mn` = 0.1, 0.1, 0.2 nm (Voxel dimensions)
- `P1mn` = 15, 15, 25 nm (PSF box dimensions)
- `fs` = 530 (FWHM scaling factor)
- `lam1`, `lam2` = 670, 518 nm (Wavelength)

The command above creates two PSF files for wavelength 670 nm and 518 nm.

- `PSF_gandy_lam670_fs530.dat`
- `PSF_gandy_lam518_fs530.dat`

## 2 Calculate *in-silico* monochrome image intensity

The *in-silico* monochrome image intensity  $I(l', m')$  is calculated using,

```
Tut1$
  siliscopy gen_mono --file dp100.gro --paramfile parameters.
  dat --psf PSF_gandy --output img100 --method slice
Tut1$
  siliscopy gen_mono --file dp2000.gro --paramfile parameters
  .dat --psf PSF_gandy --output img2000 --method slice
```

This uses the PSF files generated in the previous step (`--psf PSF_gandy`), and reads the following variables from `parameters.dat`,

- `fs = 530`
- `lam1, lam2 = 670, 518 nm`
- `lam_names1` and `lam_names2` (Particle names)
- `dlnn = 0.1, 0.1, 0.2 nm`
- `Plmn = 15, 15, 25 nm`
- `maxlen = 25, 25, 25 nm` (Maximum MS box dimensions)
- `focus_cor = 12.5 nm` (location of object focal plane)
- `opt_axis = 2` (Optical axis; 2 implies z-axis)
- `pbc = xyz` (Periodic boundary condition)
- `psf_type = 0` (Type of PSF; 0 implies depth-invariant PSF)

The command above generates image data files,

- `img100_lam670_fs530.dat`
- `img100_lam518_fs530.dat`
- `img2000_lam670_fs530.dat`
- `img2000_lam518_fs530.dat`

### 3 Generate monochrome *in-silico* microscopy images

Monochrome *In-silico* images can be generated using the following commands,

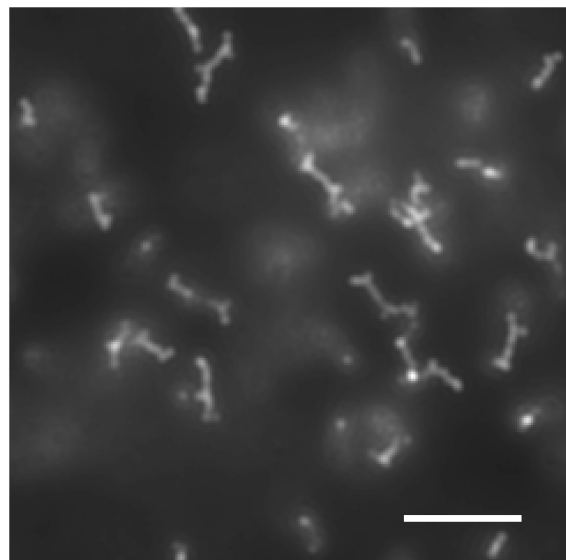
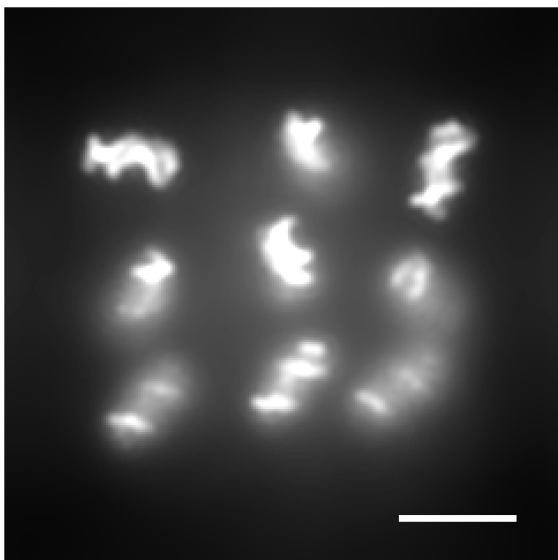
```
Tut1$ siliscopy plot --file img --paramfile parameters.dat --  
      method mono --timestep 100 --calc specific --type jpeg  
Tut1$ siliscopy plot --file img --paramfile parameters.dat --  
      method mono --timestep 2000 --calc specific --type jpeg
```

This reads the image intensity files calculated in the previous step, and reads the following variables from `parameters.dat`

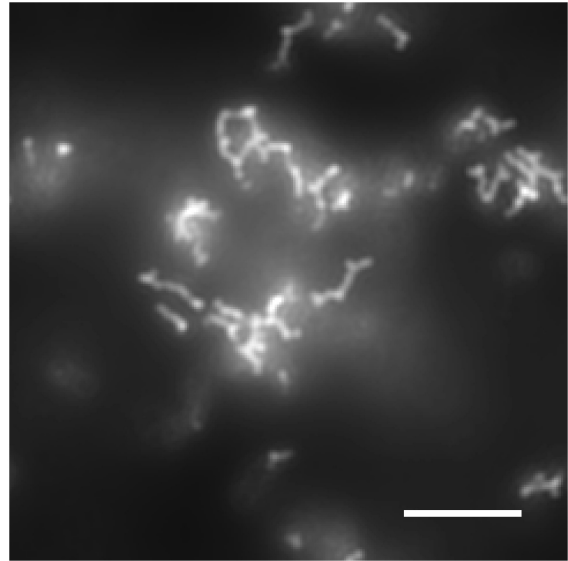
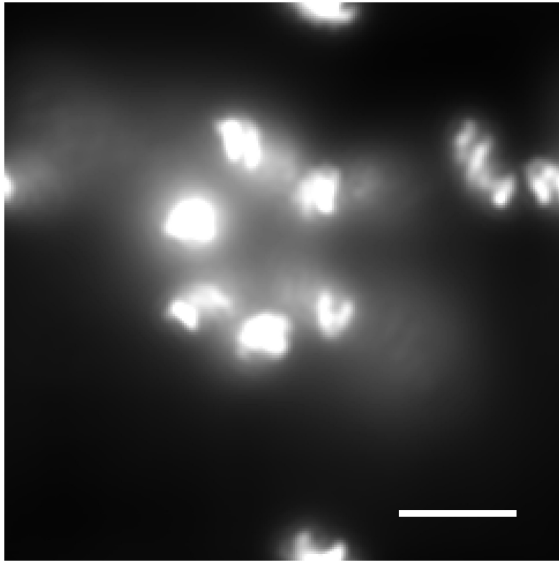
- `fs = 530`
- `T = 1` (Number of time steps used for emulating exposure)
- `scale = 5` (Size of scale bar in nm)
- `dpi = 600` (dots per inch)
- `lam1, lam2 = 670, 518`
- `lam_I0_1, lam_I0_2 = 0.13, 0.25` (Maximum intensity)
- `dlnn = 0.1, 0.1, 0.2`
- `maxlen = 25, 25, 25`
- `opt_axis = 2`

The above command generates the following images:

- `img100_lam670_fs530_T1_I0.13.jpeg`, `img100_lam518_fs530_T1_I0.25.jpeg`



- `img2000_lam670_fs530_T1_I0.13.jpeg`, `img2000_lam518_fs530_T1_I0.25.jpeg`



## 4 Generate colored *in-silico* microscopy image.

Coloured *In-silico* images can be generated using the following commands,

```
Tut1$ siliscopy plot --file img --paramfile parameters.dat --  
    method color --timestep 100 --calc specific --type jpeg  
Tut1$ siliscopy plot --file img --paramfile parameters.dat --  
    method color --timestep 2000 --calc specific --type jpeg
```

This reads the image intensity files calculated in the previous step, and reads the following variables from `parameters.dat`

- $fs = 530$
- $T = 1$
- $scale = 5$
- $dpi = 600$
- $lam1, lam2 = 670, 518$
- $lam\_IO\_1, lam\_IO\_2 = 0.13, 0.25$
- $lam\_hue1, lam\_hue2 = 255, 60$  (Hue in degrees)
- $dlnn = 0.1, 0.1, 0.2$
- $maxlen = 25, 25, 25$
- $opt\_axis = 2$

The above command generates the following images:

- `img100_fs530_T1_I_0.13_0.25.jpeg`, `img2000_fs530_T1_I_0.13_0.25.jpeg`

