



Imaging essentials checklist

- ☐ Switch the microscope on around 60 min before use to ensure temperature stability and minimize drift
- ☐ Specimen labeled with compatible dyes or fluorophores. If possible, the fluorescent sample's signal should be validated with a low magnification fluorescent microscope. Samples should not contain DAPI stain when using the Nanoimager
- ☐ Sample ready on high-precision #1.5 coverglass, multiwell slides (ONi or Ibidi), or 8-well chambers (Ibidi or Lab-Tek) are the most compatible with our stage dimensions and provide good growth conditions for cell culture. Alternatively, single coverslips, using cavity/depression slides for the addition of dSTORM buffer can be used. For some uses, coating the surface with poly-L-lysine can help with attachment.
- ☐ Basic knowledge of imaging modality to be used
- ☐ Bead slide for calibration, with 100 nm tetra spec beads
- ☐ Appropriate imaging buffer (filtered if using home-made solutions to reduce small contaminants). Sample should be covered after adding the imaging buffer to reduce exposure to oxygen.
- ☐ Avoid mounting media with high refractive index: Mounting media can prevent locking and holding focus, which will cause drift in the samples. Some autofluorescent mounting media will decrease the quality of the SMLM images.
- ☐ For live-cell imaging, please use cell culture media free from phenol red, or specific imaging buffers such as Optimem. It is recommended to add HEPES buffer media with pH7.4 to help with cell survival in the absence of CO₂
- ☐ Objective wipes
- ☐ 70-90% ethanol to clean the objective
- ☐ Oil for objective
- ☐ Magnets for stage to mount sample
- ☐ Resources to assess background levels (e.g. relevant controls)

