

# Nikon TE2000 Widefield Fluorescence and C2si Laser Scanning Confocal Microscope

## Objective Properties

Objective	NA	WD (mm)	Resolution ( $\mu\text{m}$ )	Depth of Field (mm)	Brightness	Pixel Size ( $\mu\text{m}/\text{pixel}$ )	
						Hamamatsu	Color Camera
4x PlanApo	0.20	20.00	1.53	25.0	1.0	1.653	1.197
10x PlanApo (DIC)	0.45	4.00	0.68	4.9	4.1	0.646	0.477
20x PlanApo (DIC)	0.75	1.00	0.41	1.8	7.9	0.331	0.239
40x PlanApo (DIC)	0.95	0.14	0.32	1.1	5.1	0.166	0.120
60x PlanApo VC (DIC)	1.40	0.13	0.22	0.8	10.7	0.111	0.080
100x PlanApo VC (DIC)	1.40	0.13	0.22	0.8	3.8	0.066	0.048

**NA (numerical aperture):** affects nearly everything about your image; report this along with the magnification when you publish

**WD (working distance):** how deep you can image; e.g. to image all the way through a 0.2 mm object, you need a WD > 0.2

**Resolution\*:** sizes smaller than this cannot be measured; objects closer than this distance cannot be distinguished

**Depth of Field:** the thickness of the sample that appears in focus at the same time

**Brightness:** relative measure of how much light is collected by the objective (for fluorescence only)

**Pixel size:** the size of each pixel in microns; assumes no camera binning or additional magnifying lenses

## Fluorescence Filter Properties

Fluor. Filter Set/Cube	"color"	Ex. (nm)	Em. (nm)	Example Fluorophores
#1	blue	325-375	435-485	DAPI, Hoechst
#2	green	450-490	500-550	Fluorescein, GFP, Alexa Fluor 488
#3	red	541-551	565-595	Rhodamine, propidium iodide, Alexa Fluor 546, Cy3
#4	red LP	534-556	570-640	(same as #3, but passes more light; don't use if sample has a far red dye)
#5	far red	628-672	685-735	Alexa 647, Cy5 (infrared emission; not visible to eyes - only camera)

**Note that Depth of Field, Pixel Sizes, and Filter Cube Properties DO NOT apply in Confocal Mode!**