

Matching Resolution

In digital imaging, the microscope objective lens, condenser lens, and zoom lens if any, must be “matched” such that the size of diffraction-limited Airy disk is larger than two adjacent pixels of image sensor. If Airy disk is smaller than two pixels, frequency of signal sampling is imperfect leading to significant loss of the information and the image may exhibit “**aliasing**”.

(Example) Using a Kodak KAF1400 CCD (pixel size 6.8 μm), illuminated with blue (480 nm) light and equipped with a high N.A. (1.4) objective, in epifluorescence system, the magnification yielding the maximum spatial resolution is:

$$M = (\text{“Nyquist factor”} \times \text{chip size}) / \text{“Raleigh criterion”}$$
$$= (2 \times 6.8) / (0.61 \times 0.5 / 1.4) = 62$$

Therefore, Zeiss Planapo 63x/N.A. 1.4 objective lens should provide the highest resolution possible in this system integration. In contrast, Neofluar 40x/N.A. 1.30 objective is unsatisfactory and requires a zooming lens to achieve matching resolution. If the system is transmission and uses condenser, the M must be calculated according to the Raleigh’s criterion and to the condenser must be operated in an oil-immersion mode and the diaphragm to be fully opened to achieve the highest spatial resolution.