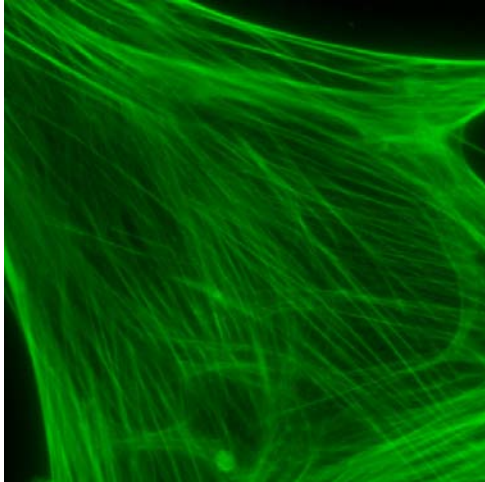


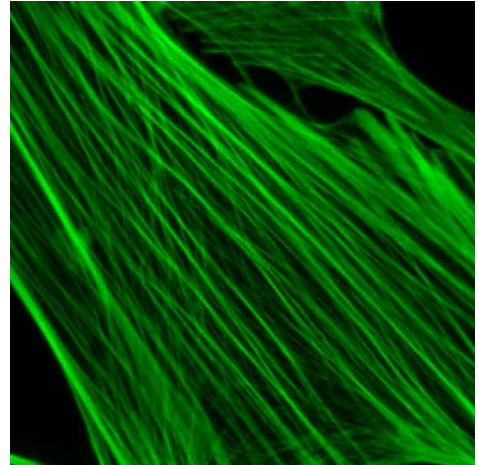
Image Quality is Often About the Little Things

Impact of Coverslip Thickness on Image Quality

New High Performance Coverslips Offered by Carl Zeiss



Coverslip #2 - ~ 200nm Thickness



Coverslip #1 ½ - ~ 170nm Thickness

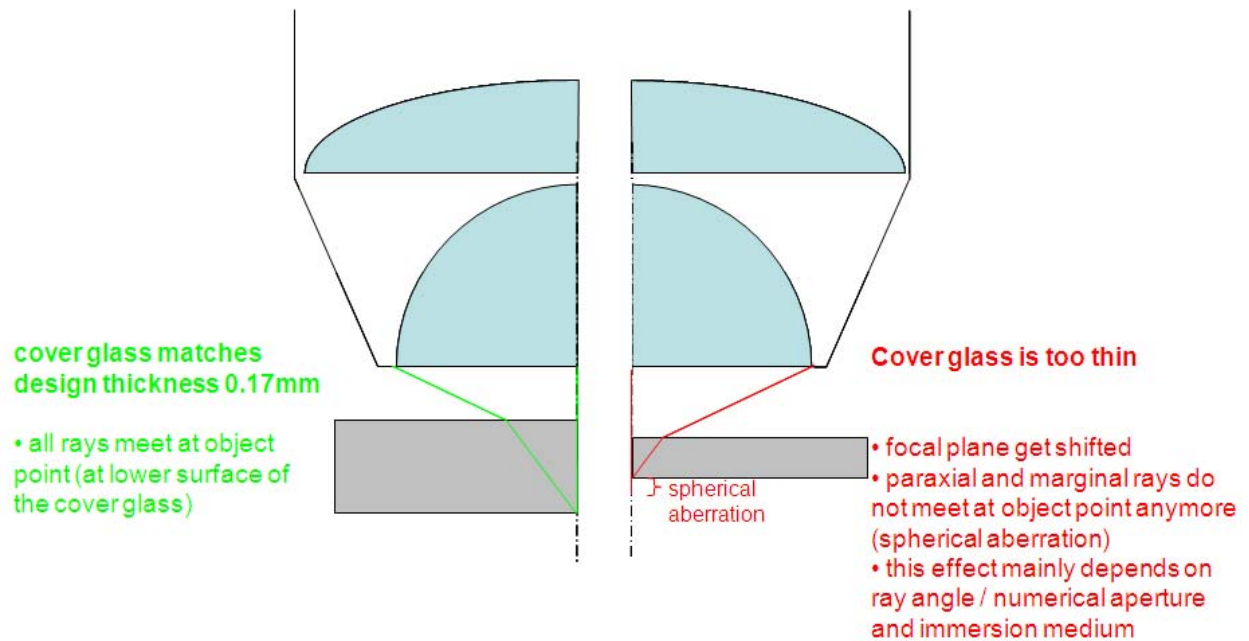
Axio Observer, Plan Apo 40x/0.95

Indian Muntjac fibroblasts stained with Alexa Fluor 488 conjugated to phalloidin

Compliments of Dr. Michael Davidson/FSU

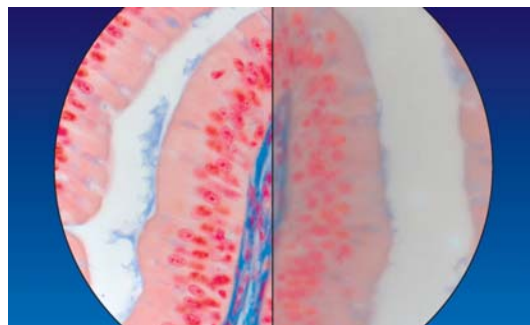
There are indeed many things that influence the performance of image quality in microscopy, many of which are overlooked relative to the enormous impact that they can have. One of those most often trivialized is the coverslip thickness. In the microscopy environment, objectives are designed assuming certain “optical conditions” present in the imaging beam path. Most objectives designed for use in high resolution biological imaging are designed based on assumptions of the “chamber window” that sits between the sample and the objective, being a coverslip of 170nm. This coverslip acts as part of the “imaging lens” and contributes significantly to the image performance.

When one varies that coverslip window, the resulting beam path change results in an increase of spherical aberration. The severity of its contribution depends upon the numerical aperture of the objective.

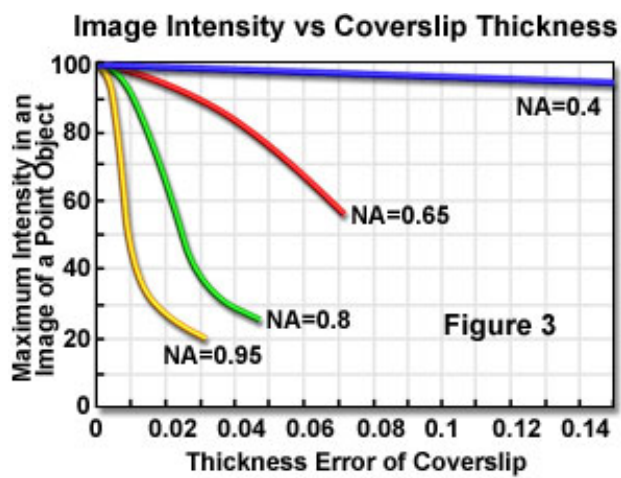
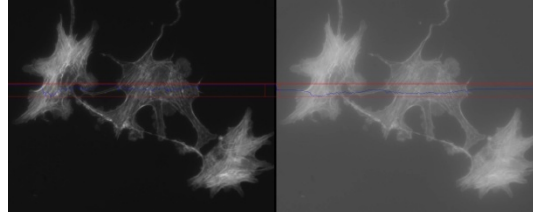


If the coverslip varies from this optimal thickness of 170 nm, the result is an increase in Image Aberrations and Severely affects the optical quality of the image. The resulting outcome:

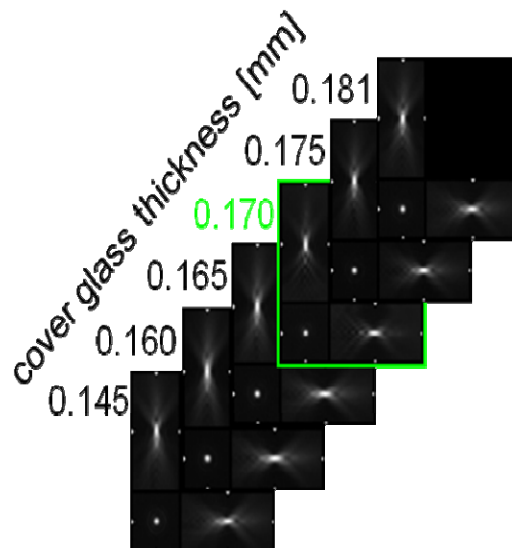
- Will cause Optical Aberrations, primarily spherical:



- Will Decrease Image Contrast and Loss of Signal



- Will Degrade the Point Spread Function:



- Will Limit the Optical Resolution

Coverslip manufacturing typically results in a widely varied range of coverslip thickness, as you can see by the following overview:

Number	Ideal Thickness	Typical Range
# 0	100 μm	80-130 μm
# 1	150 μm	130-170 μm
# 1.5	170 μm	160-190 μm
# 2	220 μm	190-250 μm

In very critical Imaging environments it has become common practice to measure the thickness of the coverslip, to allow “selection” of the required 170 nm thickness for which objectives are optimized. However, this is a very tedious process and often avoided.

Because of the critical contribution of the coverslip, Carl Zeiss has just recently announced the introduction of high tolerance coverslips, to allow users an easy solution to this problem.

- Part # **474030-9000**
- Announced March 2009
- Technical Details :
 - Size 18x18 mm²
 - Type 1 1/2 H as per ISO 8255-1 with restricted
 - thickness-related tolerance
 - D=0.17 mm +/- 0.005 mm
 - Refractive index = 1.5255 +/- 0.00015,
 - Abbe number = 56 +/- 2
 - box with 1000 Individual Cover Glasses.
- Sample of 100 Coverslips will be Included at No Charge with Every High Performance Objective Now Delivered from our factor