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# THORLABS

# N20XW-PF - August 1, 2019

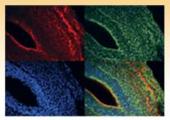
Item # N20XW-PF was discontinued on August 1, 2019. For informational purposes, this is a copy of the website content at that time and is valid only for the stated product.

# PHYSIOLOGY OBJECTIVES, WATER DIPPING OR IMMERSION

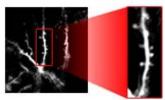
- Especially Suited for Multiphoton Imaging
- Numerical Aperture Options: 0.30 to 1.15
- Working Distance Options: 0.59 to 3.5 mm
- Designed for UV to NIR Wavelengths



N20X-PFH 1.0 NA, 2.0 mm WD



Deep Tissue Imaging of Mouse Embryo Section<sup>a</sup>



Dendridic Spine Image Collected with the N60X-NIR Objective at a Laser Wavelength of 1040 nm<sup>b</sup>

#### N16XLWD-PF 0.8 NA, 3.0 mm WD

## Hide Overview

## OVERVIEW

Thorlabs offers a selection of physiology objectives especially suited for Multiphoton Microscopy, including objectives designed for water dipping or water immersion (coverslip) setups. The high numerical apertures (NA) of these objectives help capture signal photons that are scattered through deep tissue. The long working distances (WD) and steep housing angles at the end of the objective provide access to the sample for the micromanipulators used in electrophysiology. Each objective is also designed to provide high transmission over a wide wavelength range for transmitting stimulation and emission signals. The Nikon Plan Apochromat and Plan Fluorite objectives are designed for a tube lens with focal length 200 mm, such as the ITL200 Tube Lens, whereas the Plan Fluorite Olympus objective is

designed for a tube lens with focal length 180 mm.

Thorlabs also offers the PLE153 Parfocal Length Extender for increasing the parfocal length of objectives with M25 x 0.75 threading from 60 mm to 75 mm.

- The deep tissue image featured above is courtesy of Dr. Rieko Ajima, National Cancer Institute, Frederick, MD.
- The dendritic spine image featured above is courtesy of Dr. Tobias Rose, Max Planck Institute for Neurobiology, Martinsreid, Germany.

## **Objective Lens Selection Guide**

#### Objectives

Super Apochromatic Microscope Objectives Microscopy Objectives, Dry Microscopy Objectives, Oil Immersion Physiology Objectives, Water Dipping or Immersion Long Working Distance Objectives Reflective Microscopy Objectives UV Focusing Objectives 532 nm and 1064 nm Objectives

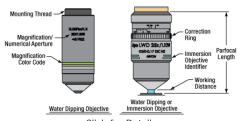
#### Scan Lenses and Tube Lenses

Scan Lenses F-Theta Scan Lenses Infinity-Corrected Tube Lenses

## Did You Know?

Multiple optical elements, including the microscope objective, tube lens, and eyepieces, together define the magnification of a system. See the *Magnification & FOV* tab to learn more.





Click for Details Examples of Water Dipping and Water Immersion Designs (See Objective Tutorial Tab for More Information About Microscope **Objective Types**)

#### Hide Objective Tutorial

# OBJECTIVE TUTORIAL

# Types of Objectives

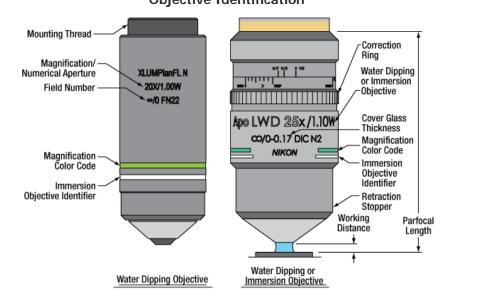
Thorlabs offers several types of objectives from Nikon and Olympus. This guide describes the features and benefits of each type of objective.

#### Water-Immersion (Coverslip) or Water-Dipping Objectives

This designation refers to the medium that should be present between the front of the objective and the specimen. Waterimmersion (coverslip) objectives are designed to work best with a drop of water and a coverslip between the objective and the specimen, while water-dipping objectives are designed to interface directly with the specimen.

#### **Plan Fluorite Objectives**

"Plan" designates that these objectives produce a flat image across the field of view. Plan



Note: These microscope objectives serve only as examples. The format of the engraved specifications will vary between objectives and manufacturers.

**Immersion Media Color Codes** 

**Magnification Color Codes** 

fluorite objectives, also referred to as plan semi-apochromats, plan fluorescence objectives, or plan fluors, are corrected for chromatic aberrations at two to four wavelengths and spherical aberrations at three to four wavelengths. Plan fluorite objectives work well for color photomicrography.

#### Plan Apochromat Objectives

"Apochromat" refers to the correction for chromatic aberration featured in the lens design. These objectives feature sophisticated designs and are corrected for chromatic corrections at four to five wavelengths and spherical aberrations at three to four wavelengths.

# **Glossary of Terms**

#### Magnification

The magnification of an objective is the lens tube focal length (L) divided by the objective's focal length (F):

# M = L/F.

The total magnification of the system is the magnification of the objective multiplied by the magnification of the evepiece or camera tube. The specified magnification on the microscope objective housing is accurate as long as the objective is used with a compatible tube lens focal length.

#### Numerical Aperture (NA)

Numerical aperture, a measure of the acceptance angle of an objective, is a dimensionless quantity. It is commonly expressed as

# **Objective Identification**

# $NA = n_i \times sin\theta_a$

where  $\theta_a$  is the maximum 1/2 acceptance angle of the objective, and  $n_i$  is the index of refraction of the immersion medium. This medium is typically air, but may also be water, oil, or other substances.

#### **Parfocal Length**

Also referred to as the parfocal distance, this is the length from the top of the objective (at the base of the mounting thread) to the bottom of the cover glass (or top of the specimen in the case of objectives that are intended to be used without a cover glass). For instances in which the parfocal length needs to be increased, parfocal length extenders are available.

#### **Working Distance**

This is the distance between the front element of the objective and the specimen, depending on the design of the objective. The cover glass thickness specification engraved on the objective designates whether a cover glass should be used.

#### **Field Number**

The field number corresponds to the size of the field of view (in millimeters) multiplied by the objective's magnification.

## FN = Field of View Diameter × Magnification

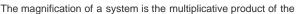
#### **Coverslip Correction and Correction Collar (Ring)**

A typical coverslip (cover glass) is designed to be 0.17 mm thick, but due to variance in the manufacturing process the actual thickness may be different. The correction collar present on select objectives is used to compensate for coverslips of different thickness by adjusting the relative position of internal optical elements. Note that many objectives do not have a variable coverslip correction (for example, an objective could be designed for use with only a standard 0.17 mm thick coverglass), in which case the objectives have no correction collar.

#### Hide Magnification & FOV

## MAGNIFICATION & FOV

# Magnification and Sample Area Calculations Magnification



magnification of each optical element in the system. Optical elements that produce magnification include objectives, camera tubes, and trinocular eyepieces, as shown in the drawing to the right. It is important to note that the magnification quoted in these products' specifications is usually only valid when all optical elements are made by the same manufacturer. If this is not the case, then the magnification of the system can still be calculated, but an effective objective magnification should be calculated first, as described below.

To adapt the examples shown here to your own microscope, please use our Magnification and FOV Calculator, which is available for download by clicking on the red button above. Note the calculator is an Excel spreadsheet that uses macros. In order to use the calculator, macros must be enabled. To enable macros, click the "Enable Content" button in the yellow message bar upon opening the file.

#### Example 1: Camera Magnification

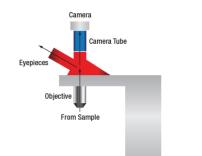
When imaging a sample with a camera, the image is magnified by the objective and the camera tube. If using a 20X Nikon objective and a 0.75X Nikon camera tube, then the image at the camera has  $20X \times 0.75X = 15X$  magnification.

#### Example 2: Trinocular Magnification

When imaging a sample through trinoculars, the image is magnified by the objective and the eyepieces in the trinoculars. If using a 20X Nikon objective and Nikon trinoculars with 10X eyepieces, then the image at the eyepieces has  $20X \times 10X = 200X$  magnification. Note that the image at the eyepieces does not pass through the camera tube, as shown by the drawing to the right.

# Using an Objective with a Microscope from a Different Manufacturer

Magnification is not a fundamental value: it is a derived value, calculated by assuming a specific tube lens focal length. Each microscope manufacturer has adopted a different focal length for their tube lens, as shown by the table to the right. Hence, when combining optical elements from different manufacturers, it is necessary to calculate an *effective* magnification for the objective, which is then used to calculate the magnification of the system.



When viewing an image with a camera, the system magnification is the product of the objective and camera tube magnifications. When viewing an image with trinoculars, the system magnification is the product of the objective and eyepiece magnifications.

Manufacturer	Tube Lens urer Focal Length	
Leica	f = 200 mm	
Mitutoyo	f = 200 mm	
Nikon	f = 200 mm	
Olympus	f = 180 mm	
Thorlabs	f = 200 mm	
Zeiss	f = 165 mm	

The rows highlighted in green denote manufacturers that do not use f = 200 mm tube lenses.



The effective magnification of an objective is given by Equation 1:

 $Effective \ Objective \ Magnification = Design \ Magnification \times \frac{f_{\textit{Tube Lens in \ Microscope}}(mm)}{f_{\textit{Design \ Tube \ Lens \ of \ Objective}}(mm)}$ 

(Eq. 1)

Here, the Design Magnification is the magnification printed on the objective, f<sub>Tube Lens in Microscope</sub> is the focal length of the tube lens in the microscope you are using, and f<sub>Design Tube Lens of Objective</sub> is the tube lens focal length that the objective manufacturer used to calculate the Design Magnification. These focal lengths are given by the table to the right.

Note that Leica, Mitutoyo, Nikon, and Thorlabs use the same tube lens focal length; if combining elements from any of these manufacturers, no conversion is needed. Once the effective objective magnification is calculated, the magnification of the system can be calculated as before.

#### Example 3: Trinocular Magnification (Different Manufacturers)

When imaging a sample through trinoculars, the image is magnified by the objective and the eyepieces in the trinoculars. This example will use a 20X Olympus objective and Nikon trinoculars with 10X eyepieces.

Following Equation 1 and the table to the right, we calculate the effective magnification of an Olympus objective in a Nikon microscope:

Effective Objective Magnification =  $20X \times \frac{200 \text{ mm}}{180 \text{ mm}} = 22.2X$ 

The effective magnification of the Olympus objective is 22.2X and the trinoculars have 10X eyepieces, so the image at the eyepieces has 22.2X × 10X = 222X magnification.

# Sample Area When Imaged on a Camera

objective magnification can be adjusted as shown in Example 3.

When imaging a sample with a camera, the dimensions of the sample area are determined by the dimensions of the camera sensor and the system magnification, as shown by Equation 2.

> $Sample Area (mm \times mm) = \frac{Camera Sensor Width (mm)}{Summer M = 1000} \times \frac{Camera Sensor Height (mm)}{Summer M = 1000} \times \frac{Cam$ System Magnification

(Eq. 2) System Magnification The camera sensor dimensions can be obtained from the manufacturer, while the system magnification is the multiplicative product of the objective magnification and the camera tube magnification (see Example 1). If needed, the

As the magnification increases, the resolution improves, but the field of view also decreases. The dependence of the field of view on magnification is shown in the schematic to the right.

## Example 4: Sample Area

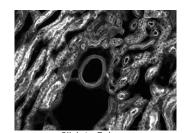
The dimensions of the camera sensor in Thorlabs' 1501M-USB Scientific Camera are 8.98 mm × 6.71 mm. If this camera is used with the Nikon objective and trinoculars from Example 1, which have a system magnification of 15X, then the image area is:

Sample Area (mm × mm) =  $\frac{8.98 \text{ mm}}{15 \text{ X}} \times \frac{6.71 \text{ mm}}{15 \text{ X}} = 599 \text{ }\mu\text{m} \times 447 \text{ }\mu\text{m}$ 

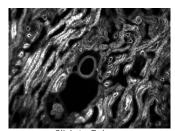
# Sample Area Examples

The images of a mouse kidney below were all acquired using the same objective and the same camera. However, the camera tubes used were different. Read from left to right, they demonstrate that decreasing the camera tube magnification enlarges the field of view at the expense of the size of the details in the image.

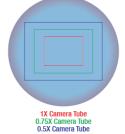




Click to Enlarge Acquired with 0.75X Camera Tube (Item # WFA4101)



Click to Enlarge Acquired with 0.5X Camera Tube (Item # WFA4102)



Sample Area When Imaged on a Camera

# Nikon Apochromat Physiology Objectives

Magnification		25X	25X	40X	40X	60X
Item #		N25X-APO- MP	N25X-APO- MP1300	N40XLWD-NIR	N40X-NIR	N60X-NIR
Manufacturer Part #		MRD77220	MRD77225	MRD77410	MRD07420	MRD07620
Numerical Aperture (NA)		1.10		1.15	0.80	1.0
Working Distance (WD)	2.0 mm		0.59 - 0.61 mm	3.5 mm	2.8 mm	
Parfocal Length	75 mm		60 mm			
Compatible Tube Lens Focal I	200 mm					
Coverslip Correction <sup>b</sup>		0 - 0.17 mm		0.15 - 0.19 mm	N/A	N/A
Immersion		Water Dipping or Water Immersion (Coverslip)		Water Immersion (Coverslip)	Water Dipping	
Wavelength Range		380 - 1050 nm	420 - 1400 nm	360 - 1100 nm	380 - 1100 nm	
Threading		M32 x 0.75		M25 x 0.75		
Temperature Range <sup>a</sup>		-18 - 60 °C (0 - 140 °F)				
Objective Case	Lid	OC2M32		OC2M25		
(Available for Purchase Separately)	Canister	OC24				

• These objectives are not recommended for use in extreme temperatures. All specifications are measured at 23 °C (73 °F).

• A coverslip correction given as a range of thicknesses indicates that the objective has a correction ring (see *Objective Tutorial* for details).

Part Number	Description	Price	Availability
N25X-APO-MP	Customer Inspired!&nbsp25X Nikon CFI APO LWD Objective, 1.10 NA, 2.0 mm WD, 380 - 1050 nm	\$29,332.76	Today
N25X-APO-MP1300	Customer Inspired!&nbsp25X Nikon CFI APO LWD Objective, 1.10 NA, 2.0 mm WD, 420 - 1400 nm	\$31,898.84	Today
N40XLWD-NIR	40X Nikon CFI APO LWD NIR Objective, 1.15 NA, 0.59 - 0.61 mm WD	\$16,875.26	Today
N40X-NIR	40X Nikon CFI APO NIR Objective, 0.80 NA, 3.5 mm WD	\$3,043.83	Today
N60X-NIR	60X Nikon CFI APO NIR Objective, 1.0 NA, 2.8 mm WD	\$4,851.14	Today

Hide Nikon Plan Fluorite Physiology Objectives

# Nikon Plan Fluorite Physiology Objectives

Magnification		10X	16X	20X	40X	
Item #		N10XW-PF	N16XLWD-PF	N20XW-PF	N40XW-PF	
Manufacturer Part #		MRH07120	MRP07220	MRF07220	MRF07420	
Numerical Aperture (NA)		0.30	0.80	0.50	0.80	
Working Distance (WD)		3.5 mm	3.0 mm	2.0 mm	2.0 mm	
Parfocal Length		60 mm	75 mm	60 mm		
Compatible Tube Lens Focal Length		200 mm				
Coverslip Correction		N/A				
Immersion	Water Dipping					
Wavelength Range		360 - 1500 nm	380 - 1100 nm	360 - 1050 nm		
Threading		M25 x 0.75	M32 x 0.75	M25 x 0.75		
Temperature Range <sup>a</sup>		18 - 60 °C (0 - 140 °F)				
Objective Case	Lid	OC2M25	OC2M32	OC2	2M25	
(Available for Purchase Separately)	Canister		OC24			

These objectives are not recommended for use in extreme temperatures. All specifications are measured at 23 °C (73 °F).

Part Number	Description	Price	Availability
N10XW-PF	Customer Inspired!&nbsp10X Nikon CFI Plan Fluorite Objective, 0.30 NA, 3.5 mm WD	\$1,570.65	Today
N16XLWD-PF	16X Nikon CFI LWD Plan Fluorite Objective, 0.80 NA, 3.0 mm WD	\$7,266.21	Today
N20XW-PF	Customer Inspired!&nbsp20X Nikon CFI Plan Fluorite Objective, 0.50 NA, 2.0 mm WD	\$2,805.11	Lead Time
N40XW-PF	Customer Inspired!&nbsp40X Nikon CFI Plan Fluorite Objective, 0.80 NA, 2.0 mm WD	\$2,933.80	Today

# Hide Olympus Plan Fluorite Physiology Objective

# **Olympus Plan Fluorite Physiology Objective**

Magnification		20X
Item #		N20X-PFH
Manufacturer Part #		1-U2B965
Numerical Aperture (NA)		1.00
Working Distance (WD)		2.00 mm
Parfocal Length		75 mm
Compatible Tube Lens Focal Length		180 mm
Coverslip Correction	rslip Correction	
Immersion		Water Dipping
Wavelength Range		400 - 900 nm
Threading	nreading	
Objective Case	Lid	OC2M25
(Available for Purchase Separately)	Canister	OC24

Part Number	Description	Price	Availability
N20X-PFH	20X Olympus XLUMPLFLN Objective, 1.00 NA, 2.0 mm WD	\$8,840.49	Today

