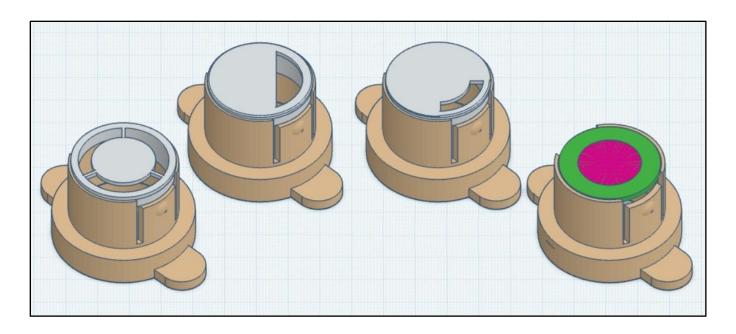
# A Darkfield/Rheinberg Accessory Kit for Olympus BH-2 Microscopes

### **Revision 4**



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Revision History		
Revision	Description of Changes	Date
1	Initial release.	August 19, 2022
2	Fixed STL files link.	December 24, 2022
3	Increased filter diameter and added grab tab to inserts. Added polarizing DF insert.	April 10, 2024
4	Added new inserts, improved darkfield webbing, and improved grab tabs on carriers.	July 29, 2025

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### Introduction

Oblique, darkfield, oblique darkfield, and Rheinberg illumination are microscopy techniques which provide optical staining of specimens under observation. These optical staining techniques are often used to enhance specimen contrast through optical methods, rather than through the application of chemical staining compounds, and are especially valuable for viewing living organisms which might otherwise be damaged or killed by the application of chemical stains. Thanks to the relative simplicity with which these techniques can be incorporated into a typical biological light microscope, and because of the stunning results that they provide, these techniques are extremely popular with amateur microscopists.

# **Oblique Illumination**

Oblique illumination is a microscopy technique that reached its peak in popularity in the late 19<sup>th</sup> century, before the existence of high N.A. objectives. Back then, oblique illumination was commonly used to highlight details in specimens that were otherwise difficult to see with the limited optics of the time. Oblique illumination works by projecting light onto the specimen under observation from an off-center (i.e., non-axial) position, creating shadowing which reveals specimen details which might otherwise be difficult to see under standard axial illumination. With the subsequent availability of high N.A. objectives towards the end of the 19<sup>th</sup> century, the technique lost much of its favor since it then became possible to see previously elusive details under standard axial illumination. However, thanks to the ease with which oblique illumination can be added to a modern biological light microscope, this technique is once again popular with amateur microscopists. Oblique illumination can be added to most microscopes by simply placing an off-center aperture stop directly beneath the aperture diaphragm within the microscope condenser, thereby obscuring on-axis light and allowing only oblique light to strike the specimen. For best results, this stop should be placed as close to the aperture diaphragm as physically possible.

### **Darkfield Illumination**

Darkfield illumination can be achieved by placing a simple stop directly beneath the aperture diaphragm within the microscope condenser, the central portion of which obscures the direct light which would normally be collected by the objective lens, and the open annulus area of which allows indirect light (which falls outside the acceptance angle of the objective lens and is therefore not collected by the objective lens) to

illuminate the specimen. When no specimen is present on the stage, this condition produces a completely dark visual field since all direct (i.e., zero order) light is obscured, leaving only indirect light which is not collected by the objective lens. When a specimen is then placed on the stage, some of the indirect light that strikes the specimen will be scattered/diffracted such that it enters the collection angle of the objective lens and is collected to form the final image. The result is a brightly illuminated specimen against a completely dark background.

### **Oblique Darkfield Illumination**

Oblique darkfield illumination is a combination of oblique illumination and darkfield illumination, wherein the open annulus of a darkfield stop is modified such that only a portion of the annulus is open (such as a 90° sector), thereby forcing the indirect light which illuminates the specimen to come from a single, off-axis direction, rather than from all sides of the specimen. This method produces darkfield images with the characteristic shadowing provided by oblique lighting.

### **Rheinberg Illumination**

Rheinberg illumination works in a similar fashion to darkfield, except that the darkfield stop is replaced by a concentric, bi-colored filter to create the Rheinberg effect. In a Rheinberg filter, the central disk of the darkfield stop (which, in darkfield, blocks the direct light from entering the objective lens) is replaced by an optical filter material of some color, to impart this color onto the direct (i.e., zero order) light, rather than blocking it. The outer annulus portion of the darkfield stop, through which the indirect light that falls outside the acceptance area of the objective lens comes through, is replaced by an optical filter material of some contrasting color to the central disk, to impart this contrasting color onto the indirect light. Since the indirect light is not collected by the objective, the color of the indirect light does not show up in the visual field when there is no specimen on the stage, leaving a background of the color of the central disk of the filter. When a specimen is then placed on the stage, some of the indirect light that strikes the specimen will be scattered/diffracted such that it enters the collection angle of the objective lens and is collected to form the The result is a brightly illuminated final image. specimen which is primarily colored by the annulus portion of the filter, in stark contrast to the background, which is colored by the central disk.

### **Adding Stops and Filters to the BH-2**

Ideally, darkfield stops, oblique stops, oblique darkfield stops, and Rheinberg filters should be located at the

the microscope condenser, but this is of course not physically possible for an add-on accessory for a conventional condenser. Given this limitation, best results will be obtained when the stops or filters are placed as close to the condenser aperture diaphragm as possible. The collar-mount condensers used on many microscopes are constructed such that a filter carrier, which is present on the bottom of the condenser, is sufficiently close to the aperture diaphragm that stops and filters placed within this carrier provide good results. In comparison, microscopes which use dovetailmounted condensers (such as the Olympus BH-2 and Nikon scopes from the same era), are notoriously difficult for the amateur to equip for oblique, darkfield, oblique darkfield, or Rheinberg illumination. reason for this difficulty is that the condensers on these scopes are constructed in such a way that it is not possible for a filter carrier on the bottom of the condenser to position a stop or filter sufficiently close to the aperture diaphragm to provide acceptable results. In order to use a simple stop or filter to obtain oblique, darkfield, oblique darkfield, or Rheinberg on these scopes, a carrier of some sort is necessary which can be placed up into the bottom bore of the condenser to position the stop or filter very close to the aperture diaphragm within.

same optical plane as the aperture diaphragm within

# **Scope of this Document**

This document describes the BH2-DFR Accessory Kit, which is a collection of 3D-printed plastic parts that allows oblique, darkfield, oblique darkfield, or Rheinberg illumination to be easily added to any BH-2 microscope equipped with a BH2-CD or BH2-AAC condenser. The fabrication and usage of the BH2-DFR Accessory Kit are described within this document.

### **Versions of the BH2-DFR**

There are two versions of the BH2-DFR which can be made from the components in the BH2-DFR Accessory Kit. The BH2-DFR-CD version was designed to be used with the BH2-CD condenser (the non-corrected Abbe condenser commonly found on BHT and BHTU scopes, which has an N.A. of 1.25), and the BH2-DFR-AAC version, which was designed to be used with the (the BH2-AAC condenser aplanatic achromatic condenser commonly found on BHS scopes, which has an N.A. of 1.4). Although the two versions are similar, the BH2-DFR-AAC version places the stops or filters a bit deeper into the body of the condenser than the BH2-DFR-CD version, since this is where the aperture diaphragm in the BH2-AAC is located.

### **Cross-Compatibility**

The BH2-DFR-CD version of the BH2-DFR can be freely used with either BH2-CD or BH2-AAC condensers. When the BH2-DFR-CD version is used in a BH2-AAC condenser, the stop inserts will be positioned a bit further away from the aperture diaphragm than would be the case if the recommended BH2-DFR-AAC version were used. This means that the results obtained using the BH2-DFR-CD version in the BH2-AAC condenser will, at least in theory, be inferior to what the BH2-DFR-AAC version would provide (although the difference will be very minimal).

The BH2-DFR-AAC version should never be used in the BH2-CD condenser, as damage to the internal iris mechanism within the condenser may result.

# **An Alternate Solution for Oblique Illumination**

Although the BH2-DFR Accessory Kit does provide oblique illumination capabilities for the Olympus BH-2, a better solution for oblique illumination is the BH2-OBL Accessory Kit (see *An Oblique Accessory Kit for Olympus BH-2 Microscopes*), which was specifically designed to provide oblique illumination for the Olympus BH-2. The BH2-OBL is superior for oblique illumination in that it includes a provision to allow the user to easily change the physical positioning of the oblique aperture while viewing the specimen.

# The BH2-DFR Accessory Kit

The BH2-DFR Accessory Kit consists of a collection of simple 3D-printed plastic parts, as described in the sections below. These parts can be easily made using virtually any low-cost 3D FDM printer.

### The Insert Carriers and Filter Carriers

Both the BH2-DFR-CD and BH2-DFR-AAC versions of the BH2-DFR include an insert carrier for use with the darkfield, oblique, oblique darkfield, and custom/filter inserts in the kit, as well as two filter carriers (shallow and deep) for use with Rheinberg filters of differing thicknesses. The shallow filter carriers accept filters up to 1.5mm in thickness, whereas the deep carriers accept filters up to 3.0mm in thickness. BH2-DFR-CD carriers (Figure 1) can be distinguished from the BH2-DFR-AAC carriers (Figure 2) by the presence of a slight step-down in the outer diameter of the upper portion of the BH2-DFR-AAC carriers (Figure 2). The filter carriers can be distinguished from the insert carriers by the presence of a filter support ledge within the upper portion of the filter carriers (Figure 1), which supports standard 32mm diameter filters. Be sure to always use the appropriate filter carrier that matches the thickness of the filters you wish to use, otherwise the iris mechanism of the condenser could be damaged if the filter protrudes too far from the upper end of the carrier.

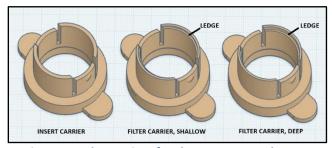


Figure 1 – The carriers for the BH2-CD condenser.

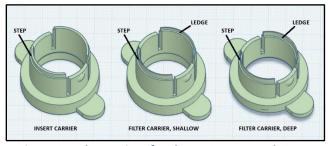


Figure 2 - The carriers for the BH2-AAC condenser.

Since the BH2-DFR-CD and BH2-DFR-AAC versions of the BH2-DFR are similar in appearance, only the BH2-DFR-CD version will be illustrated throughout the remainder of this document.

### The Complete Set of Inserts

The complete set of inserts, which are compatible with both the BH2-DFR-CD and BH2-DFR-AAC insert carriers, are shown in **Figure 3**. These inserts provide oblique, darkfield, oblique darkfield, and Rheinberg capabilities to the BH2-DFR.

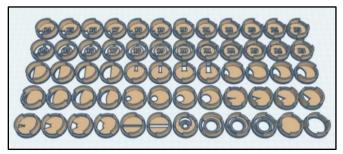


Figure 3 – The complete set of BH2-DFR inserts.

### The Top Filter Insert

The BH2-DFR Accessory Kit includes a top filter insert (Figure 4) which can accommodate filters from 28.0mm to 28.5mm in diameter. This insert can be used to place custom filters or stops into the upper opening of the BH2-DFR. When using this insert with a stop or filter, the stop or filter must be secured in the central recess using some sort of adhesive, since gravity will not hold the disk in place on the underside of the insert.

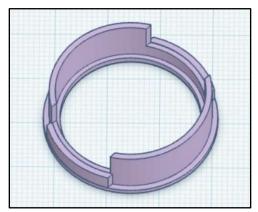


Figure 4 - The "top filter" insert.

### **The Bottom Filter Insert**

The BH2-DFR Accessory Kit includes a bottom filter insert (Figure 5) which can be used to place filters into the lower opening of the BH2-DFR insert carriers. The bottom filter insert can accommodate filters from 28.0mm to 28.5mm in diameter.

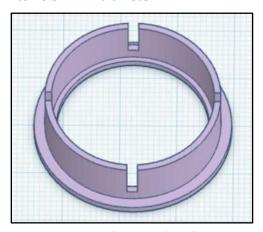


Figure 5 - the "bottom filter" insert.

### The Hybrid Filter Inserts

The BH2-DFR Accessory Kit includes four hybrid inserts (Figure 6) with center recesses made to accommodate filters, for adding polarizing or color filters to the optical setup for oblique, darkfield, and oblique darkfield. These hybrid inserts include one oblique insert (left), which accepts filters from 12.2mm to 14.3mm in diameter, two oblique darkfield inserts (second and third from left), and one darkfield insert (right), which accept filters from 17.5mm to 18.0mm in diameter. When using these inserts with filters, the filters must be secured in place in the central recess using some sort of adhesive, since gravity will not hold the filters in place on the underside of the inserts.

A high-extinction linear polarizer placed within the center recesses will allow for polarizing oblique, polarizing darkfield, or polarizing oblique darkfield, wherein the user can control the degree to which darkfield is produced by rotating a second high-

extinction linear polarizer placed over the field lens (below the condenser) to obtain the desired effect. This setup allows the user to quickly select between brightfield, darkfield, or anything in between, by simply rotating the lower polarizer, and without ever having to touch the condenser.

A color filter placed within the center recesses will produce modified oblique (left), modified oblique darkfield (second and third from left), or modified darkfield (right), wherein the background of the visual field is tinted the color of the filter, rather than appearing black.

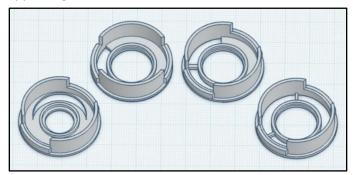


Figure 6 – The "hybrid filter" inserts.

### **The Closed Custom Insert**

The BH2-DFR Accessory Kit includes a closed filter insert (Figure 7) for custom applications. This insert can be used to make custom stops for specific applications not covered by the existing inserts, by simply drilling or cutting custom apertures into the face of the insert, or by importing the STL file for the closed insert into appropriate 3D modeling software and editing within, to obtain the desired custom printed stops.

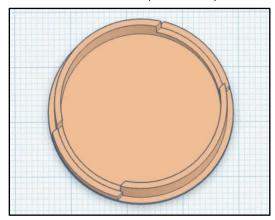


Figure 7 – The "closed custom" insert.

### **The Darkfield Inserts**

The BH2-DFR Accessory Kit includes a comprehensive collection of darkfield inserts with central stops ranging in diameter from 14mm to 25mm, in 1mm increments (Figure 8). These inserts provide darkfield capabilities for objective lenses up to 40X magnification.

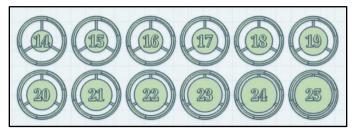


Figure 8 - The "darkfield" inserts.

# **The Oblique Darkfield Inserts**

The BH2-DFR Accessory Kit includes a comprehensive collection of oblique darkfield inserts, which provide single-quadrant illumination, with central stops ranging in diameter from 14mm to 25mm, in 1mm increments (Figure 9). These inserts provide oblique darkfield capabilities for objective lenses up to 40X magnification.

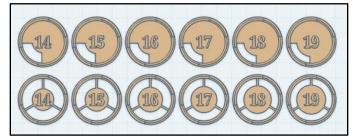


Figure 9 – The "oblique darkfield" inserts.

# **The Oblique Inserts**

The BH2-DFR Accessory Kit includes a comprehensive collection of oblique inserts (Figure 10) for various forms of oblique lighting.

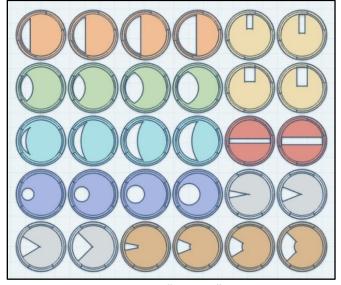


Figure 10 – The "oblique" inserts.

### **BH2-DFR Configurations**

The BH2-DFR Accessory Kit can be configured to provide any of the four basic illumination modes (darkfield, oblique, oblique darkfield, or Rheinberg) as shown in

**Figure 11**. The procedures to set up the BH2-DRF for these modes are described in the sections below.

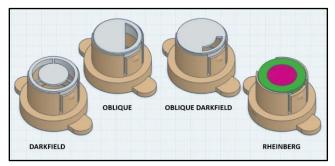


Figure 11 – The various configurations of the BH2-DFR.

### Oblique, Darkfield, and Oblique Darkfield

The BH2-DFR Accessory Kit can be configured to provide oblique, darkfield, or oblique darkfield illumination by simply placing the desired insert type into either the BH2-DFL-CD insert carrier (Figure 1, left) or BH2-DFR-AAC insert carrier (Figure 2, left), as appropriate. When installing inserts into the insert carriers, be sure to align the inserts such that the two alignment tabs on the insert (Figure 12) seat into the clearance spaces above the spring tabs of the carrier (Figure 13), so that the insert drops fully into the carrier and does not pose a risk of damage to the iris diaphragm mechanism within the condenser.

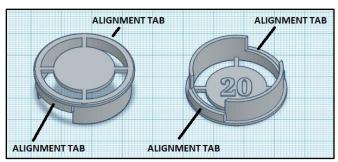


Figure 12 – The alignment tabs of the inserts.

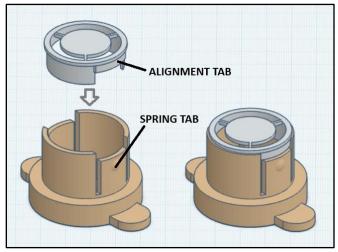


Figure 13 – Proper alignment of the inserts in the carrier.

### Using a Bottom Filter in the Insert Carrier

A 28mm to 28.5mm diameter filter may be installed in the bottom of either the BH2-DFR-CD or BH2-DFR-AAC insert carriers, by using the bottom filter insert (Figure 5). Simply place the desired filter into the bottom filter insert (Figure 14) and press the bottom filter insert, with the filter in place, into the bottom of the insert carrier (Figure 15).

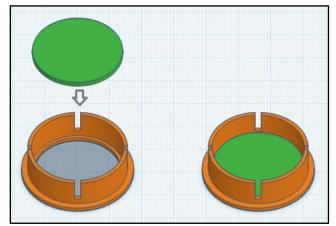


Figure 14 - Placing a filter into the bottom filter insert.

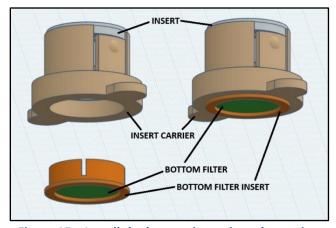


Figure 15 – Install the bottom insert into the carrier.

### **Rheinberg Illumination**

The BH2-DFR Accessory Kit can be configured to provide Rheinberg Illumination by simply placing a 32mm diameter Rheinberg filter (not included in the BH2-DFR kit) into the appropriate BH2-DFR-CD filter carrier (Figure 1, middle and right), or the appropriate BH2-DFR-AC filter carrier (Figure 2, middle and right), making sure to match the thickness of the filter to the depth of the filter ledge within the filter carrier (shallow or deep), to prevent damage to the condenser.

### **Download the STL Files**

Before you can print the various component parts of the specific version of the BH2-DFR that you wish to make, you must first obtain the appropriate 3D model STL files for those specific parts.

### **STL Files for the Insert Carrier and Filter Carriers**

The STL files for the three carriers for the BH2-DFR-CD version of the BH2-DFR are listed in **Table 1**.

Table 1 – STL Files for the BH2-DFR-CD Carriers			
File Location: BH2-DFR_STL_Files.zip /Carriers			
Images of these carriers are shown in Figure 1.			
Insert Carrier	Insert Carrier Left Insert_Carrier_CD.stl		
Filter Carrier, Shallow Middle Filter_Carrier_Shallow_CD.st		Filter_Carrier_Shallow_CD.stl	
Filter Carrier, Deep Right Filter_Carrier_Deep_CD.stl			

The STL files for the three carriers for the BH2-DFR-AAC version of the BH2-DFR are listed in **Table 2**.

Table 2 – STL Files for the BH2-DFR-AAC Carriers			
File Location: BH2-DFR_STL_Files.zip /Carriers			
Images of these carriers are shown in Figure 2.			
Insert Carrier	sert Carrier Left Insert_Carrier_AAC.stl		
Filter Carrier, Shallow Middle Filter_Carrier_Shallo		Filter_Carrier_Shallow_ AAC.stl	
Filter Carrier, Deep Right Filter_Carrier_Deep_ AAC.stl		Filter_Carrier_Deep_ AAC.stl	

# **STL Files for the Top and Bottom Filter Inserts**

The STL files for the top filter insert and the bottom filter insert of the BH2-DFR are listed in Table 3.

Table 3 – STL Files for the BH2-DFR Custom and Filter Inserts			
File Location: BH2-DFR_STL_Files.zip /Filters			
Images of these inserts are shown in Figure 4 and Figure 5.			
Top Filter Insert Figure 4 Top_Filter.stl			
Bottom Filter Insert	Figure 5	Bottom_Filter.stl	

# **STL Files for the Custom and Hybrid Inserts**

The STL files for the four hybrid filter inserts and the custom closed filter insert of the BH2-DFR are listed in Table 4.

Table 4 – STL Files for the BH2-DFR Custom and Filter Inserts				
File Location: BH2-DFR_STL_Files.zip /Filters				
Images of thes	Images of these inserts are shown in Figure 6.			
Oblique, w/ Filter	left	Oblique_Filter.stl		
Oblique Darkfield 1 w/ filter	2 <sup>nd</sup> from left	Oblique_DF_Filter_1.stl		
Oblique Darkfield 2 w/ filter	3rd from left	Oblique_DF_Filter_2.stl		
Darkfield w/ filter right		DF_Filter.stl		
Image of this insert is shown in Figure 7.				
Closed Custom Insert Custom_Closed_Insert.stl				

# **STL Files for the Darkfield Inserts**

The STL files for the darkfield inserts of the BH2-DFR are listed in **Table 5**.

Table 5 – STL Files for the BH2-DFR Darkfield Inserts			
File Locatio	File Location: BH2-DFR_STL_Files.zip /Darkfield		
Images are these inserts are shown in Figure 8.			
Darkfield Insert, 14mm Top row, left Darkfield_14.stl			
Darkfield Insert, 15mm	Top row, 2nd from left	Darkfield_15.stl	

Table 5 – STL Files for the BH2-DFR Darkfield Inserts		
Darkfield Insert, 16mm	Top row, 3rd from left	Darkfield_16.stl
Darkfield Insert, 17mm	Top row, 4th from left	Darkfield_17.stl
Darkfield Insert, 18mm	Top row, 5th from left	Darkfield_18.stl
Darkfield Insert, 19mm	Top row, right	Darkfield_19.stl
Darkfield Insert, 20mm	Bottom row, left	Darkfield_20.stl
Darkfield Insert, 21mm	Bottom row, 2nd from left	Darkfield_21.stl
Darkfield Insert, 22mm	Bottom row, 3rd from left	Darkfield_22.stl
Darkfield Insert, 23mm	Bottom row, 4th from left	Darkfield_23.stl
Darkfield Insert, 24mm	Bottom row, 5th from left	Darkfield_24.stl
Darkfield Insert, 25mm	Bottom row, right	Darkfield_25.stl

# **STL Files for the Oblique Darkfield Inserts**

The STL files for the oblique darkfield inserts are listed in **Table 6**.

Table 6 – STL Files for the BH2-DFR Oblique Darkfield Inserts			
File Location: BH2-DFR_STL_Files.zip /Oblique_Darkfield			
Images o	Images of these inserts are shown in Figure 9.		
Oblique DF Insert, 14mm	Top row, left	Oblique_Darkfield_14.stl	
Oblique DF Insert, 15mm	Top row, 2nd from left	Oblique_Darkfield_15.stl	
Oblique DF Insert, 16mm	Top row, 3rd from left	Oblique_Darkfield_16.stl	
Oblique DF Insert, 17mm	Top row, 4th from left	Oblique_Darkfield_17.stl	
Oblique DF Insert, 18mm	Top row, 5th from left	Oblique_Darkfield_18.stl	
Oblique DF Insert, 19mm	Top row, right	Oblique_Darkfield_19.stl	
Oblique DF Insert, 20mm	Bottom row, left	Oblique_Darkfield_20.stl	
Oblique DF Insert, 21mm	Bottom row, 2nd from left	Oblique_Darkfield_21.stl	
Oblique DF Insert, 22mm	Bottom row, 3rd from left	Oblique_Darkfield_22.stl	
Oblique DF Insert, 23mm	Bottom row, 4th from left	Oblique_Darkfield_23.stl	
Oblique DF Insert, 24mm	Bottom row, 5th from left	Oblique_Darkfield_24.stl	
Oblique DF Insert, 25mm	Bottom row, right	Oblique_Darkfield_25.stl	

# **STL Files for the Oblique Inserts**

The STL files for the oblique inserts of the BH2-DFR are listed in **Table 7**.

Table 7 – STL Files for the BH2-DFR Oblique Inserts				
File Location: BH2-DFR_STL_Files.zip /Oblique				
Images of ti	Images of these inserts are shown in Figure 10.			
Sliced Circle 1	Row 1, left	Sliced_Circle_1.stl		
Sliced Circle 3	Row 1, 2 <sup>nd</sup> from left	Sliced_Circle_2.stl		
Sliced Circle 3	Row 1, 3 <sup>rd</sup> from left	Sliced_Circle_3.stl		
Sliced Circle 4	Row 1, 4 <sup>th</sup> from left	Sliced_Circle_4.stl		
Notch 1	Row 1, 5 <sup>th</sup> from left	Notched_1.stl		
Notch 2	Row 1, right	Notched_2.stl		
Football 1	Row 2, left	Football_1.stl		
Football 2	Row 2, 2 <sup>nd</sup> from left	Football_2.stl		
Football 3	Row 2, 3 <sup>rd</sup> from left	Football_3.stl		
Football 4	Row 2, 4 <sup>th</sup> from left	Football_4.stl		
Notch 3	Row 2, 5 <sup>th</sup> from left	Notched_3.stl		
Notch 4	Row 2, right	Notched_4.stl		
Crescent 1	Row 3, left	Crescent_1.stl		
Crescent 2	Row 3, 2 <sup>nd</sup> from left	Crescent_2.stl		
Crescent 3	Row 3, 3 <sup>rd</sup> from left	Crescent_3.stl		

Table 7 – STL Files for the BH2-DFR Oblique Inserts				
File Location: BH2-DFR_STL_Files.zip /Oblique				
Images of t	Images of these inserts are shown in Figure 10.			
Crescent 4	Row 3, 4 <sup>th</sup> from left	Crescent_4.stl		
Slit 1	Row 3, 5 <sup>th</sup> from left	Slit_1.stl		
Slit 2	Row 3, right	Slit_2.stl		
Circle 1	Row 4, left	Circle_1.stl		
Circle 2	Row 4, 2 <sup>nd</sup> from left	Circle_2.stl		
Circle 3	Row 4, 3 <sup>rd</sup> from left	Circle_3.stl		
Circle 4	Row 4, 4 <sup>th</sup> from left	Circle_4.stl		
Pie 1	Row 4, 5 <sup>th</sup> from left	Pie_Slice_1.stl		
Pie 2	Row 4, right	Pie_Slice_2.stl		
Pie 3	Row 5, left	Pie_Slice_3.stl		
Pie 4	Row 5, 2 <sup>nd</sup> from left	Pie_Slice_4.stl		
Sector 1	Row 5, 3 <sup>rd</sup> from left	Sector_1.stl		
Sector 2	Row 5, 4 <sup>th</sup> from left	Sector_2.stl		
Sector 3	Row 5, 5 <sup>th</sup> from left	Sector_3.stl		
Sector 4	Row 5, right	Sector_4.stl		

All of the STL files listed in **Table 1** through **Table 7** are included in the **BH2-DFR\_STL\_Files.zip** file, which is available for download at the following Google Drive location:

https://drive.google.com/drive/folders/1YmmqtSaPhHaZnf 6zRctKjqlMN-LYIQj4?usp=share\_link

### Slice the STL Files

With the necessary STL files in hand, the next step is to use *slicer* software to process the STL files, to produce GCODE files which are compatible with your specific model of 3D printer. While the exact procedure for using the slicer software is beyond the scope of this document, there are some specific slicing parameters which are recommended. Configure the slicer to create GCODE for a standard 0.4mm nozzle size and 0.2mm layer height, using a 3-layer shell thickness with 20% or greater infill. The various component parts of the BH2-DFR Accessory Kit were designed such that no supports, rafts, nor brims are needed to successfully print the parts.

# **Print the Component Parts**

Once you have created the necessary GCODE files from the STL files, the component parts can then be printed on your 3D printer. Before printing, make sure you have a standard 0.4mm nozzle installed on your printer and that the printer has been properly configured to use this nozzle. The parts may be printed using standard PLA filament (this is acceptable for most applications), but if you will be using extremely intense lighting, you should consider a filament with a higher melting point than PLA, such as PETG, ABS, ASA, or Nylon. If possible, print the insert carriers and filter

carriers in something other than PLA, as PLA tends to creep when exposed to continuous stress. This creep means that the spring tabs which hold the carriers in place may deform over time, causing the grip within the condenser to loosen. If PLA is your only option, just reprint a new carrier whenever one gets too loose to use. Whichever type of filament you choose, be sure to use black (or better still, spend a few extra dollars and get a flat-black filament, if available) to reduce the potential for light reflections within the device. Once you have printed the necessary parts, use an X-Acto knife (or similar) with a sharp blade to trim away any bumps, ridges, burrs, or stringing.

### **Test-Fit the Carriers in the Condensers**

The BH2-DFR-AAC version of the BH2-DFR is not compatible with the BH2-CD condenser. Do not insert the BH2-DFR-AAC carriers into a BH2-CD condenser, otherwise physical damage to the iris mechanism within the BH2-CD condenser may result.

Carefully align and press the appropriate insert carrier (BH2-DFR-CD or BH2-DFR-AAC) into the bottom bore of the applicable condenser (BH2-CD or BH2-AAC), until the flange on the carrier seats against the bottom surface of the mounting dovetail ring on the condenser (Figure 16, carrier shown in tan for visibility), and then remove it from the bore, taking notice of the insertion and extraction forces required to do so. If necessary, use fine-grit sandpaper or emery cloth to smooth any rough exterior features of the carrier, until an acceptable fit within the condenser bore has been achieved. Be sure to thoroughly remove any sanding dust before re-inserting the carrier back into the Once the appropriate BH2-FDR insert condenser. carrier has been properly fitted to the condenser, repeat the test-fitting procedure with the appropriate shallow and deep filter carriers, using the same condenser.

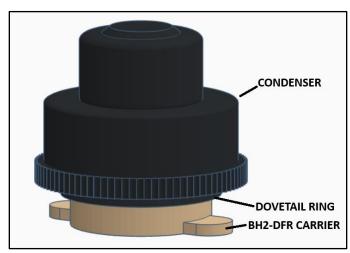


Figure 16 - The BH2-DFR fitted into a BH2 condenser

### How to Use the BH2-DFR

The procedure for using the BH2-DFR is described in the sections below.

### **A Few Words of Caution**

Due to the close proximity of the inserts of the BH2-DFR to the iris mechanism within the condensers, it is critical that the inserts remain properly engaged with their respective carriers whenever the BH2-DFR is installed, to prevent damage to the fragile iris mechanism within the condenser. It is strongly recommended that once you find a specific configuration that you wish to use, create a dedicated carrier for this configuration and use cyanoacrylate adhesive (or some other suitable adhesive) to securely bond the insert into the carrier. This will eliminate the risk of damage to the condenser caused by a displaced insert.

Do not adjust the aperture-control ring of the condenser while the BH2-DFR is installed in the condenser. Always leave the aperture diaphragm in the wide-open position while the BH2-DFR is installed.

### Setup Microscope for Köhler Illumination

Before installing the BH2-DFR onto your microscope, perform a routine Köhler setup using the objective lens you wish to use, to ensure that the condenser is axially centered within the illumination path of the microscope.

# **Configure the BH2-DFR for the Desired Mode**

As described in the *BH2-DFR Configurations* section of this document, place the desired insert or Rheinberg filter into the upper end of the appropriate BH2-DFR carrier, and (if applicable) install the bottom filter carrier, with the filter present, into the lower end of the insert carrier.

### Install the BH2-DFR onto the Microscope

The BH2-DFR-AAC version should never be used in the BH2-CD condenser, as damage to the internal iris mechanism within the BH2-CD condenser could result.

Use the procedure described below to install the BH2-DFR onto your microscope.

- Use the condenser focus knob to lower the microscope condenser as far as it will go.
- Loosen the condenser locking thumbscrew on the substage assembly of the microscope stand and carefully remove the condenser from the substage assembly.
- Adjust the aperture-control ring of the condenser to set the aperture diaphragm to the wide-open position.
- With the desired insert or filter in place in the appropriate carrier (and with the bottom filter in place, if applicable), carefully press the BH2-DFR into the bottom bore of the condenser until the

- flange on the carrier contacts the circular mounting dovetail of the condenser (Figure 16).
- Reinstall the condenser with the BH2-DFR fitted onto the substage assembly of the microscope stand and snug the condenser locking thumbscrew to secure it in place.
- Use the condenser-focus knob to return the condenser to its proper operating position.

To prevent damage to the condenser, do not rotate the aperture-control ring while the BH2-DFR is installed in the condenser.

# **Tips for Using the BH2-DFR**

There are many online sites geared towards amateur microscopy, and these sites have a great many references that describe various methods, tips, and tricks for using oblique, darkfield, oblique darkfield, and Rheinberg illumination. That information is readily available and as such will not be repeated here. Listed below are a few basic things to get you started when using the BH2-DFR on your BH-2 scope.

- Proper condenser centering is critical for achieving optimal results. The best way to center the condenser is to perform a routine Köhler setup on the objective you will be using before inserting the BH2-DFR into the condenser bore.
- Proper condenser height is critical for achieving optimal results.
   Don't be afraid to experiment with the condenser focus knob to see what setting gives the best results.
- Always pre-set the condenser aperture diaphragm to the wideopen setting before installing the BH2-DFR and leave it at that setting while the BH2-DFR is installed in the condenser.
- The minimum required diameter of the central patch of darkfield and oblique darkfield stops (and the diameter of the central disk of Rheinberg filters) depends on the N.A. of the objective lens that will be used. The higher the N.A. of the objective, the larger the diameter needs to be.
- A simple way to determine the proper diameter of the central patch stop, as well as the central disk for Rheinberg filters, is to set the N.A. of the condenser to match the N.A. of the objective lens with which it will be used, and then remove the condenser from the scope and measure the resulting opening in the iris mechanism within the condenser. In practice, you should use a somewhat larger stop than this minimum diameter (perhaps 20% larger), to account for imperfections in the height and centering of the condenser, as well as imperfections in the centering of the patch stop or filter within the condenser.
- The BH2-DFR can provide great results with almost any objective lens up to 20X magnification. Acceptable performance at 40X can usually be achieved with careful setup of condenser height and centering, along with the proper selection of patch stop diameter.
- For darkfield or oblique darkfield with a 40X objective, use an A40, EA40, or DPlan40 objective, as these optics have a modest N.A. and will therefore give you the best results. Don't waste your time with higher performance 40X objectives, as the NA of these make it all but impossible to achieve darkfield.
- The BH2-DFR cannot provide darkfield, oblique darkfield, or Rheinberg illumination with 100X objective lenses, as the N.A. of such objectives is too high for simple stops to provide acceptable

results. A special oil-wetted darkfield condenser (the BH2-DCW), along with a 100X objective lens with an integral aperture diaphragm (to reduce the N.A. of the objective) is necessary to obtain darkfield from 100X objectives.

- Darkfield is brutal. A single speck of dust or debris at the specimen plane (i.e., on the slide, cover slip, or near the specimen) will light up brightly against the dark background and severely detract from the final image. No matter how well you clean things, you will never be able to eliminate all of the hot spots. So, what about those perfect darkfield images you see in all the articles? How did they get those? Photoshop is your friend.
- While an undersized darkfield stop may not provide complete darkfield, it can provide a noticeable reduction in the intensity of the background light, which may be desirable in some situations.
- The custom insert can be used to make custom stops, by drilling
  or cutting the desired illuminating aperture(s) into the face of the
  insert. Better still, import the STL file for the custom insert into
  Tinkercad or Fusion 360 and add the desired features to the
  model before printing the part.
- If you find yourself using the same two or three inserts or filters
  over and over in your BH2-DFR, do yourself a favor and print a
  separate carrier for each of these, and then use cyanoacrylate
  adhesive (or some other suitable adhesive) to glue each insert or
  filter into its own dedicated carrier. The convenience that this
  provides (as well as safety for the condenser) makes it well worth
  the effort.
- If you're lucky enough to have a second condenser on hand, consider keeping one equipped with your favorite BH2-DFR darkfield setup (a 20mm stop is a good choice), and use it as your dedicated darkfield condenser. Use the other condenser for straight brightfield work.
- Wow, there are a lot of parts here, aren't there? If you just want darkfield, but don't want to waste a lot of time printing unneeded parts or trying out things that don't work, print the appropriate insert carrier for your condenser, along with the 20mm darkfield insert. Glue these together and you will have all you need for good darkfield on your BH-2 scope. And that's only two parts!
- Why waste time with different sizes of darkfield stops? Why not
  just use a stop with a large diameter all the time? The larger the
  stop diameter, the less illumination your specimens will receive.
   So, while the stop needs to be big enough to reliably achieve the
  darkfield effect, any larger than this needlessly detracts from the
  maximum image brightness.
- Cut a disk of linearly polarized film (the higher the extinction ratio, the better) and glue it into the center recess of the hybrid filter inserts. Now, whenever you use these inserts, you can place a 45mm linearly polarized filter in the filter recess of the field lens under the condenser, and when oriented perpendicular to the polarizing axis of the upper disk, you'll get darkfield. Rotate it 90° and you'll get brightfield. Now isn't this way more convenient than continually fussing with the condenser, to go from brightfield to darkfield? Note that the presence of the lower polarizer reduces the maximum available light intensity available for darkfield by 50%. Note also that if you remove the filter from the field lens, you will get brightfield, but with a reduction in the intensity of the background visual field of 50%, due of the presence of the polarizer in the hybrid insert.
- Do you use both the BH2-CD and BH2-AAC condensers? If so, to make things easier on yourself, just print the parts for the BH2-CD

condenser and use those in either condenser. Practically speaking, you'll never see the difference. Since it is not recommended to use the BH2-AAC parts in the BH2-CD condenser, if you use both condenser types, it's just safer to keep only the -CD version around.

### **How To Contact the Author**

Please feel free to direct any questions or comments regarding this document (or BH-2 microscopes in general) to the author as listed on the cover page of this document.