OXFORD INSTRUMENTS ANDOR

BC43

3.1 rev 04 September 2024

User Guide

For model: BC433

andor.oxinst.com

Section 1 - Safety and Warning Information

Caution
 **PLEASE READ THIS INFORMATION FIRST BEFORE USING YOUR PRODUCT** **USE OF CONTROLS OR ADJUSTMENTS OR PERFORMANCE OF PROCEDURES OTHER THAN THOSE SPECIFIED HEREIN MAY RESULT IN HAZARDOUS RADIATION EXPOSURE.**

1. If the equipment is used in a manner not specified by Andor, the protection provided by the equipment may be impaired.
2. Do not position this product so that it is difficult to operate the mains disconnecting device. See "Emergency Mains Disconnection" on page 99.
3. Before using the system, please follow and adhere to all warnings, and safety, manual handling, and operating instructions located either on the product, or in this manual.
4. Keep this manual in a safe place for future reference.
5. Users must be authorised and trained personnel only; otherwise, this may result in personal injury, and/or equipment damage and impaired system performance.
6. There are few user-serviceable parts inside the product and the enclosure must only be opened to perform the specified operations detailed in "Operation" on page 99 and "Maintenance and User Service Procedures" on page 126.
7. IEC Technical Document IEC TR 60825-14 recommends the presence of a Laser Safety Officer (LSO); however, national guidelines should be referred to.
8. This equipment has not been designed and manufactured for the medical diagnosis of patients.

9. Do not attempt to bypass any safety interlocks. They are provided to comply with the safety requirements of various regulatory agencies and must be employed to protect the operator.
10. Protective earth is an integral part of the protection against electric shock in this product and is provided via the earth pin of the external power supply. Ensure that this is plugged into the building earth system via the mains socket. Do not tamper with any of the earthing measures.
11. Any AC/DC Power Supply used with this product must meet the requirements specified in "Electrical Power Specifications" on page 221
12. Only use a mains cord certified to the safety regulations for your region and suitably rated for the mains supply specified in "Electrical Power Specifications" on page 221.
13. Make sure the power supply cord is located so that it will not be subject to damage. If replacement of the detachable power supply cord is required, ensure replacement is of same type and rating.
14. While running an experiment, keep room temperature as stable as possible.
15. Performance of the system may be adversely affected by rapidly changing environmental conditions or operation outside of the operating conditions specified in "Technical Specifications" on page 215
16. Electromagnetic Compatibility: This is a Class A product. In a domestic environment this product may cause electromagnetic interference, in which case the user may be required to take adequate measures.
17. This product has been designed and tested to perform successfully in a normal (basic) electromagnetic environment, e.g. a typical life science test laboratory, as per the EU EMC Directive. It is not designed to operate in a harsh electromagnetic environment, e.g. close to the following equipment: EMI/RFI generators, electrostatic field generators, electromagnetic or radioactive devices, plasma sources, arc welders, x-ray instruments, intense pulsed sources, or other similar sources of high energy fields whose emissions are not within the normal range expected under the EU EMC Directive.
18. Please note that this product is not designed to provide protection from ionising radiation. Any customer using this product in such an application should provide their own protection.

19. This product is a precision scientific instrument containing fragile components. Always handle it with care.
20. Do not store or place liquids on the product.
21. If spillage occurs on the product, switch off power immediately, and wipe off with a dry, lint-free cloth.
22. If any ingress of liquids has occurred or is suspected, unplug the mains cables, do not use, see "Cleaning and Decontamination Procedures" on page 126
23. Do not expose the product to extreme hot or cold temperatures.
24. Do not expose the product to open flames.
25. Do not allow objects to fall on the product.

Caution

CAUTION: FLASHING LIGHT SOURCES: PLEASE BE AWARE THAT THIS PRODUCT CONTAINS LIGHT SOURCES THAT RAPIDLY FLASH BETWEEN OFF AND ON. THIS MAY AFFECT SOMEONE WITH EPILEPSY WHO SUFFERS FROM PHOTOSENSITIVITY. TAKE APPROPRIATE PRECAUTIONS IN CONJUNCTION WITH YOUR OCCUPATIONAL HEALTH & SAFETY MANAGER.

1.1 Revision History

Version	Released	Description
1.0	1st November 2021	Initial release.
1.1	19th November 2021	Updated troubleshooting section. Updated Sample Levelling procedure, replacing the Front Access Panel Door Push to Release Mechanism procedure. Updated Front Door Replacement procedure and "China RoHS Hazardous Substances Declaration" on page 227.
1.2	13th December 2021	Updated accessories code table. Updated Sample Levelling procedure, updated Front Access Panel Door Push to Release Mechanism procedure.
1.3	10th January 2022	Added new section Recommended Sample Substrates, Updated FSL description, section 3.7.3. Updated troubleshooting section.
1.4	14th March 2022	Added new section "PC Workstation Operating System Notes and IT / Networking" on page 58. Added new troubleshooting entries.
1.5	17th May 2022	Added new troubleshooting section to "Troubleshooting" on page 201.
1.6	22nd July 2022	Updated hyperlink to Fusion guide for BC43. Added new entry to troubleshooting table. Added note on Objectives table (Section 1.4.1). Added extra note on ensuring BC43 LED is solid blue before starting Fusion (Section 3.5). Added additional mechanical drawings to show BC43 with lid open ("Mechanical Drawings" on page 222). Updated ROHS table.
1.7	30th November 2022	Updated objectives section in accessories table. Added new entries to the troubleshooting section regarding image focus, and montage protocol. Added new entry to troubleshooting table regarding FSL calibration. Added new entry to troubleshooting table regarding connection of the Spacemouse joystick to the PC. Updated RoHS Printed Circuit Board Assemblies and Screw Locks. Updated Replacing the XY Stage procedure. Updated levelling the sample holder procedure to account for lock screws. Added new section on Windows Operating System Updates.
1.8	13th January 2023	Added "PC Workstation Operating System Notes and IT / Networking" on page 58. New sections "Creating Additional Windows Accounts" on page 61 and "Changing the PC Workstation Name or Media Access Control (MAC) Address" on page 61
1.9	15th February 2023	Added new Troubleshooting entry - Fusion reports no valid license. Directed to section 3.4.3
2.0	13th March 2023	3.3.2 Added Spacemouse buttons connection instructions and a troubleshooting entry. "BC43 and PC Workstation Setup" on page 54 new PC box replaced original PC. Earlier setup in Appendix C and USB 3.0 for joystick instruction. Added 2 troubleshooting entries regarding unlisted objective lenses. Added Consumables section to "Accessories" on page 43. Added 2 oils in. "Replacing the XY Stage" on page 152 Step 8 Warning and extra step added.

Version	Released	Description
2.1	18th April 2023	Changes to the "BC43 and PC Workstation Setup" on page 54. New PC tower image and changed instructions. "Technical Specifications" on page 215 Changed objectives available to 100x magnification
2.2	29th June 2023	Troubleshooting entries regarding connecting to Fusion software, PC powering ON, blank live preview and blocked confocal disk.
2.3	30th June 2023	Added "Chamber Self Calibration" on page 96
2.4	16th October 2023	Appendix A, "BC43 Laser Power Specifications" on page 219, added words 'back aperture' in title. Updated section "Assembly" on page 54, added new Appendix "Connection Diagrams for Previous PC Models" on page 224. Added new section "Adjusting the Multiwell Plate Holder" on page 61. Updated "Accessories" on page 43.
2.5	23rd October 2023	Updated section "Assembly" on page 54.
3.0	25th July 2024	Added new sections on "Advanced Maintenance Procedures" on page 166. Updated Japan and China office addresses. Added new section on "Anti-virus software" on page 61. Added "Model overview of Benchtop Microscopes Family. " on page 34Updated "BC43 Laser Power Specifications" on page 219. Updated "BC43 and PC Workstation Setup" on page 54 and "Connection Diagrams for Previous PC Models" on page 224
3.1	04 September 2024	Added new section "Patent Information" on page 40

1.2 Updates to the Manual

Changes are periodically made to the product, and these will be incorporated into new editions of the manual. Please check for new releases of the manual at: andor.oxinst.com/downloads. If you find an issue in this manual, please contact your customer support representative with a description of the issue.

1.3 Unpacking Information

Carefully unpack the unit and retain the packaging materials to transport or return equipment if required:

- If the equipment appears damaged in any way, return it to sales outlet in its original packaging.
- No responsibility for damage arising from the use of non-approved packaging will be accepted.
- Ensure all items and accessories specified at the time of ordering and as detailed on the packing list are present: if any items are missing, please contact your sales representative.

1.4 Laser Safety Information

- This symbol on the rear panel advises that it is mandatory to read this user guide before using this product.

- As defined in IEC 60825-1, this product is Class 2 when the lid is open, which means that it is low power and eye protection is normally afforded by natural aversion behaviour, i.e. you close your eye lids and look way before it can hurt your eyes. Thus, it is defined as "safe momentarily" for eyes, and safe for diffuse reflections, skin, and fire.
- Note that the beam emitted vertically upwards from the objective is stopped from propagating further by the aluminium Transmitted Light Turret.
- As the eye aversion response protects against Class 2 radiation, you are warned not to stare into the beam.
- To understand the relative safety of this product, a typical Class 2 product is a laser pointer.
- Although the technical classification of the product is Class 2, it is hard to get laser radiation into your eyes due to the beam path. You would need to use a reflective object to do so or place your head in an unusual and awkward position.
- Therefore, take special care when using reflective or metal objects near the laser radiation. This product allows Class 2 laser radiation to be used when the lid is open to provide a bright enough light source to enable users to position their samples.
- When the lid is down, this product is Class 1, i.e. completely safe.
- All lasers are CW (Continuous Wave), are generated using laser diodes, are confined to a very narrow bandwidth and have very narrow, low-divergence beams.
- Please see "BC43 Laser Power Specifications" on page 219.

- The instrument should not be disassembled more than is required in the procedures in "Operation" on page 99 and "Maintenance and User Service Procedures" on page 126, otherwise you endanger yourself and possibly others as the laser radiation can be at Class 3B levels inside normally inaccessible parts of the product during operation, which is hazardous and can cause permanent eye damage.

1.4.1 Laser Safety Protection Measures

The following protection measures are used in the product to ensure that you only have access to safe Class 1 laser radiation when the lid is closed or Class 2 "momentarily safe" laser radiation when the lid is open, in accordance with the international product laser safety standard IEC 60825-1 and U.S. CDRH Regulations 21 CFR 1040.10 and 1040.11:

1.4.2 Protective Housing

Any laser radiation above Class 1 AEL (Accessible Emission Limit) is housed in either an aluminium housing or inside optical fibre, except in the case of laser radiation above the microscope objective. During operation, this will be below the Class 2 AEL when the laser radiation is accessible for sample positioning (i.e. with the lid open). However, when the lid is closed the laser radiation inside can be more than the Class 2 AEL, but it is inaccessible due to the lid and the beam is stopped from propagating further by the aluminium Transmitted Light Turret.

1.4.3 Safety Interlocks

Safety interlocks are automatic devices associated with each portion of the protective housing of a laser product that are normally used to prevent human access to laser radiation when that portion of the protective housing is removed, opened or displaced. However, in this product they are used to reduce Class 3B radiation to Class 2 when the lid is open, to reduce the risk to what is necessary to position your samples accurately. The following safety features should be noted regarding the interlocks used in this product:

- All interlocks are redundant, i.e. there are two switches in each circuit so that if one fails short, then the other will continue to protect you
- The switches are certified for safety use by UL
- Interlocks are provided in both the lid and the front door, and are used to activate the samplepositioning mode which keeps the laser radiation within

the Class 2 AEL

- The sample-positioning mode is based on placing an interlocked beam attenuator in the laser beam path to reduce the laser radiation below Class 2 AEL to allow safe laser radiation to be used to position the sample
- When the lid or front door is opened, the laser emission will stop for a very brief moment until this attenuator is in place and will then start emitting at up to Class 2 AEL levels
- After 5 minutes of being open, the laser will stop emitting and will not resume until the lid is closed and opened again; this is a de-risking feature as sample positioning should not require longer than this

1.4.4 Manufacturing Tests

The product has been tested during manufacturing to ensure that the accessible laser radiation is less than Class 2 AEL in all operating modes.

1.4.5 Fault Analysis

The product has been extensively analysed at the component level to ensure that single faults cannot cause exposure to laser radiation above Class 2 AEL. Product safety standards assume that it is not reasonably foreseeable that multiple faults could occur to cause a hazard and that further faults could not accumulate after a single fault without the problem being noticed, i.e. the majority of single faults cause the product to fail and others cannot accumulate with subsequent faults to cause a hazard. Moreover, nearly all single faults are highly unlikely, and most are extremely unlikely.

1.5 Laser Class Overview

1.5.1 Laser Class Nominal Power Limits

AEL (Accessible Emission Limit) means the "maximum accessible emission permitted within a particular class" (IEC 60825-1 3rd Edition Section 3.3). For the visible spectrum (400 – 700 nm) based on IEC 60825-1 Section 4.3 for ≥ 0.25 sec exposure as measured under Condition 3 (which is a 7 mm limiting aperture at 100 mm from the objective that simulates an eye's pupil at the most likely minimum distance), the AELs can be described simply as follows:

- Class 1 begins at 39 µW for 400 nm, rising to 390 µW at 500 nm and remaining at 390 µW up to 700 nm for ≥ 100 sec exposures.
- Class 2 is up to 1 mW for ≥ 0.25 sec exposures for simple, low-divergence laser beams called 'small sources'. However, the laser output of this product consists of many tiny laser beams which make it a 'complex source' and the higher-divergence makes it also an 'extended source', so the power output is allowed to be as high as 2.9 mW before it reaches the Class 2 AEL. However, as can be seen on the label we have placed a maximum limit of 1.3 mW for all versions of this product when the lid is open, but the actual 100% Condition 3 power output values for this version can be found in "BC43 Laser Power Specifications" on page 219.
- Class 3B is 500 mW for ≥ 0.25 sec.
- Class 4 is anything above Class 3B.

1.5.2 Laser Class Practical Differences

The practical differences between the main classes are as follows as defined in IEC 60825-1 Annex C and Table F.2:

Class	Overview*	Eye	Diffuse Reflections	Skin	Fire	Hazard Detail
1	Safe under reasonably foreseeable conditions	Safe	Safe	Safe	Safe	Laser products that are safe during use, including long-term direct intrabeam viewing, even when exposure occurs while using telescopic optics
2	Low power; eye protection normally afforded by aversion & active responses	Safe momentarily, due to natural aversion behaviour, i.e. you will close your eye lids and look way	Safe	Safe	Safe	Laser products that emit visible radiation that are safe for momentary exposures but can be hazardous for deliberate staring into the beam. The time base of 0.25 s is inherent in the definition of the class and presumption is that there is very low risk of injury for momentary exposures that are somewhat longer
3R‡	Direct intrabeam viewing may be hazardous	Not considered intrinsically safe	Safe	Safe	Safe	Not considered intrinsically safe, the risk is limited because – unintentional exposures would rarely reflect worst-case conditions of (e.g.) beam alignment with a large pupil and worst-case accommodation with the entire beam

Class	Overview*	Eye	Diffuse Reflections	Skin	Fire	Hazard Detail
						energy entering the eye, – of the inherent reduction factor (safety margin) in the MPE, – of natural aversion behaviour for exposure to bright light for the case of visible radiation and by the response to heating of the cornea for far infrared radiation. The risk of injury increases with exposure duration, and exposure may be hazardous for ocular exposure under worst-case conditions or for intentional direct intrabeam viewing.
3B	Direct intrabeam viewing normally hazardous, i.e. direct viewing of beam with eye	Hazardous	Normally safe	Lasers which approach the Class 3B AEL may produce minor skin injuries. However, this is only likely if the beam has a small diameter or is focussed	Risk of igniting flammable materials for same lasers mentioned under "Skin	Laser products that are normally hazardous when intrabeam ocular exposure occurs (i.e. within the NOHD) including accidental short time exposure

Class	Overview*	Eye	Diffuse Reflections	Skin	Fire	Hazard Detail
4	High power: diffuse reflections may be hazardous	Hazardous	May be hazardous	Hazardous	Often represent a fire hazard	Everything above Class 3B
‡ CDRH Class IIIa is almost identical to Class 3R and 21CFR § 1040.10 (b) (8) describes the hazard as "depending upon the irradiance, either an acute intrabeam viewing hazard or chronic viewing hazard, and an acute viewing hazard if viewed directly with optical instruments"						

1.6 Laser Aperture Location

The Laser Aperture, where the laser radiation is emitted, is the microscope objective, or, in the absence of an objective, the socket where the objective is attached. The yellow arrow in the figure below shows where the laser radiation is emitted and that it is stopped from propagating further by the Transmitted Light Turret.

Figure 1: The Laser Aperture, where the laser radiation is emitted, is the microscope objective, or, in the absence of an objective, the socket where the objective is attached

1.7 Operational Laser Access

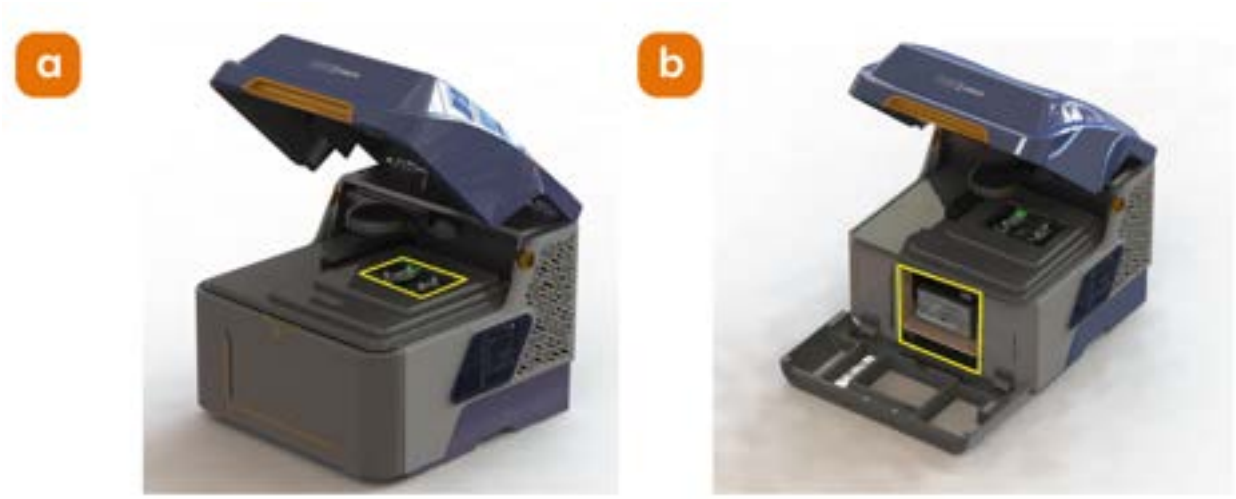

Figure 2: Operational laser access. a) Sample holder and b) Focus Seek and Lock opening.

1.8 Laser Class 2 Warning Labels

1.8.1 Laser Product Classification

- The following label classifies this product as a Class 2 Laser Product based on the maximum accessible laser radiation
- See the information at the beginning of this Laser Safety Information section
- The power values on this label are intended as maximum values for classification purposes based on IEC 60825-1 Condition 3 taking into account future versions of this product. The actual values for your version of the product can be found in "BC43 Laser Power Specifications" on page 219

Figure 3: Label classifies this product as a Class 2 Laser Product.

1.8.2 Access Panel Labels

- The following labels are located adjacent to the top lid on the front door panel and on the inside of the open front door panel
- These indicate that when opened these panels allow access to Class 2 laser radiation
- See the information at the beginning of this Laser Safety Information section

Figure 4: Label located adjacent to the top lid on the front door panel.

1.9 Laser Class 3B Warning Labels

There are a number of labels on the rear and bottom exterior of the unit, and in the interior of the unit that indicate access panels or fibre connections that give access to hazardous Class 3B laser radiation as shown below. These are strictly for service staff only and not users, and must not be opened. In some cases, the access actually means that laser will be emitted from an aperture, so these are identified as laser apertures in the labels below.

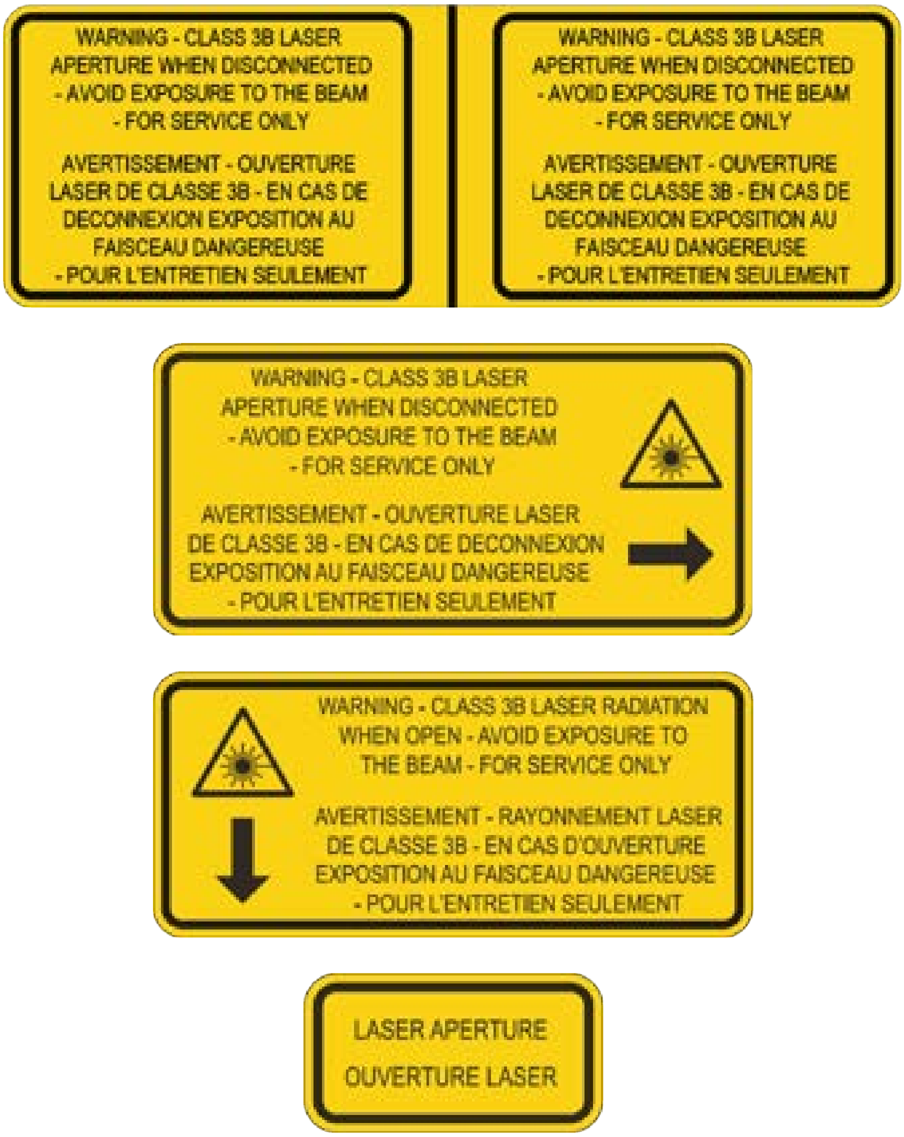

Figure 5: Labels on the rear and bottom exterior of the unit.

1.10 Other Safety Information

1.10.1 Multi-person Lift

Figure 6: Lift warning label on BC43.

If this unit is being lifted or moved, at least three people are required to do this. Follow good practice for manual handling, especially the guidance provided by your employer. See "Unpacking BC43" on page 51.

1.11 Mechanical Safety

This product has been risk-assessed in accordance with the EU Machinery Directive, esp. all moving parts. The following risks should be noted, and any moving parts not mentioned have been assessed and are not deemed a risk.

The following symbol signifies a finger trap hazard and is only found on the Focus Seek and Lock Module as the other potential finger trap hazards would require an unusual set of circumstances to occur. Please see "Risk No 3: Focus Seek and Lock Module Replacement" on page 27.

Figure 7: Finger crush warning label on BC43.

1.12 Risk No 1: Finger Crush Between Y Stage and Rear Chassis

Warning
The user is advised to be aware of this hazard, which could crush a finger in an unusual set of circumstances

Description of Potential Hazard

In normal use the Y plate can travel as indicated below. As the Y plate approaches the rear chassis the resultant gap can reduce to 1.0 mm, which is less than the minimum 25 mm gap allowed for a finger in Table 13 of IEC 61010-1.

A finger placed in the gap would be crushed if the Y plate moved fully back to the rear chassis.

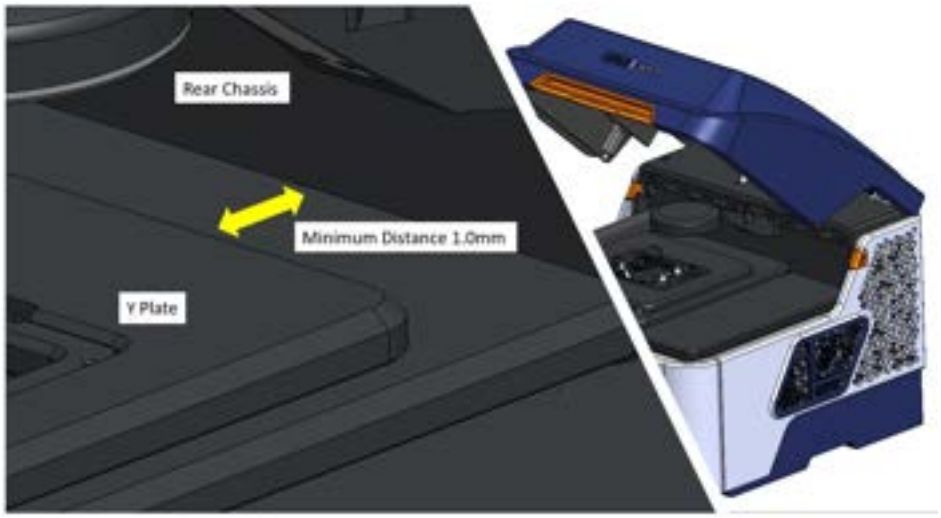

Figure 8: Schematic indicating the finger crush hazard.

Mitigating Factors

- It would be a highly unusual scenario for a user to put their finger in this position
- It would be even more unusual for the right circumstances to occur, e.g. get a protocol going in software and then move to get your hand in place to get hurt

- The Y plate travels relatively slowly (6 mm/sec)
- The motor is audible

Risk Assessment per IEC 61010-1

Severity: Moderate M (sufficient to bruise or scratch a body part)

Probability of Exposure: E1 (exposure is not intended during normal use)

Possibility of Avoidance: P1 (possibility of avoidance)

Conclusion

As per IEC 61010-1, the level of risk requires no action, and the hazard is such as to make reduction or elimination impractical to implement.

1.13 Risk No 2: Insertion of Finger into Drive Box / Touch Rotating Parts

Warning
The user is advised to be aware of this hazard, which could crush a finger in an unusual set of circumstances

Description of Potential Hazard

In normal use, the Y plate travels as indicated while the drive box remains stationary. Depending on the position of the Y plate, there may be an accessible aperture on the side of the drive box.

A finger placed in the accessible aperture would be crushed as the Y plate moves fully back. The gap can reduce to zero.

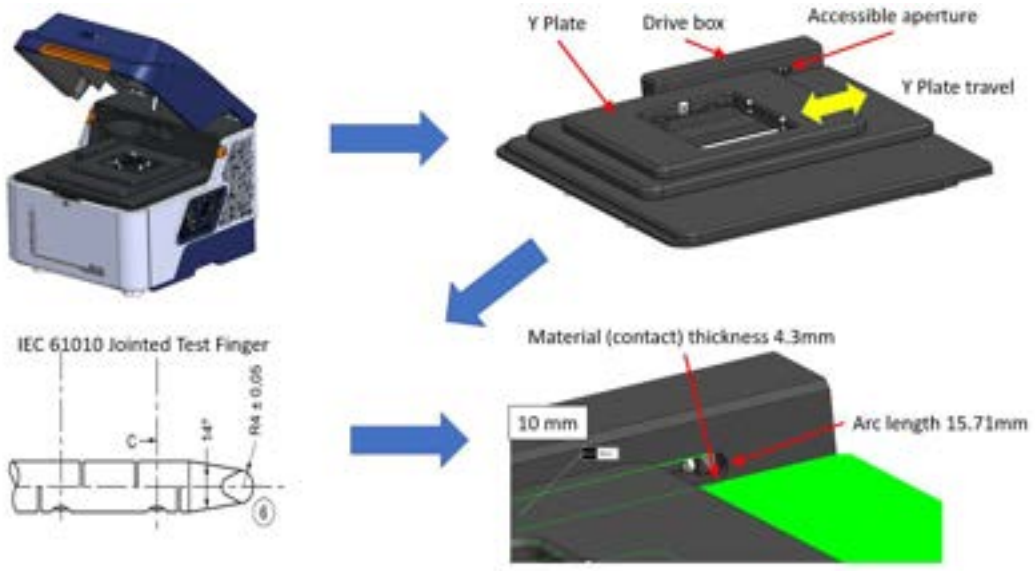

Figure 9: Schematic indicating the finger crush hazard.

Mitigating Factors

- It would be a highly unusual scenario for a user to put their finger in this position.
- It would be even more unusual for the right circumstances to occur, e.g. get a protocol going in software and then move to get your hand in place to get

hurt.

- The Y plate travels relatively slowly (6 mm/sec).
- The motor is audible.

Risk Assessment per IEC 61010-1

Severity: Moderate M (sufficient to bruise or scratch a body part)

Probability of Exposure: E1 (exposure is not intended during normal use)

Possibility of Avoidance: P1 (possibility of avoidance)

Conclusion

As per IEC 61010-1, the level of risk requires no action, and the hazard is such as to make reduction or elimination impractical to implement.

1.14 Risk No 3: Focus Seek and Lock Module Replacement

Warning
Ensure the instrument is powered down and disconnected from the mains supply before this activity to eliminate the risk of crushing a finger in unusual circumstances as described below

Description of Activity

It may be necessary for the user to remove the Focus Seek and Lock for cleaning during the lifetime of the product. The electrical connection is achieved via a blind mate connection and the procedure is as follows:

- Release the single fixing screw
- Grip the Focus Seek and Lock body's finger grips and remove it from the instrument

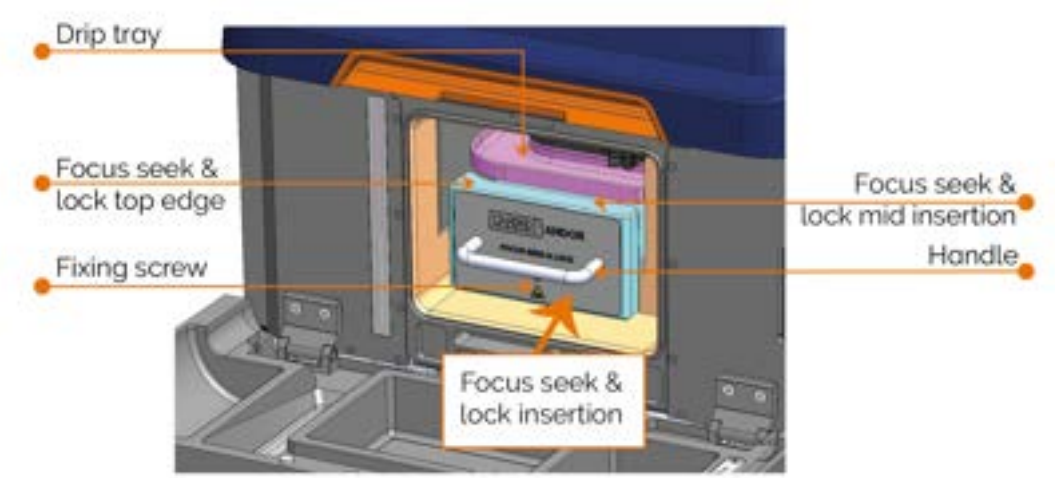

Figure 10: Schematic indicating the finger crushing hazard.

Description of Potential Hazard

If power is re-established while someone is replacing the Focus Seek and Lock and a finger is placed over/beyond the barrier and under the drip tray, the Z stage could potentially home and crush a finger.

Mitigating Factors

- There is no risk during normal maintenance activity
- This is contrary to guidance in the user guide and is thus misuse
- It is an unnatural scenario for a user to put their finger in this position
- Another person would have to establish power while the first person is replacing the Focus Seek and Lock, which is not a very reasonable scenario

Risk Assessment

Although possible, this is a very improbable set of circumstances and thus there is negligible risk as it is not reasonably foreseeable.

Conclusion

The hazard is such as to make reduction or elimination impractical to implement, but the risk is negligible.

1.15 Risk No 4: XY Stage Replacement

Warning
Ensure the instrument is powered down and disconnected from the mains supply before this activity to eliminate the risk of crushing a finger in unusual circumstances as described below

Description of Activity

It may be necessary for the user to remove/replace the XY Stage during the lifetime of the product. The XY Stage's electrical connection is achieved via a blind mate connection and the procedure is as follows:

- Remove or reposition the objective lenses to enable tool access to the rear fixing screw
- Release the 3 fixing screws: 2 at the front and 1 at the rear
- The XY Stage can now be removed from the main body of the instrument

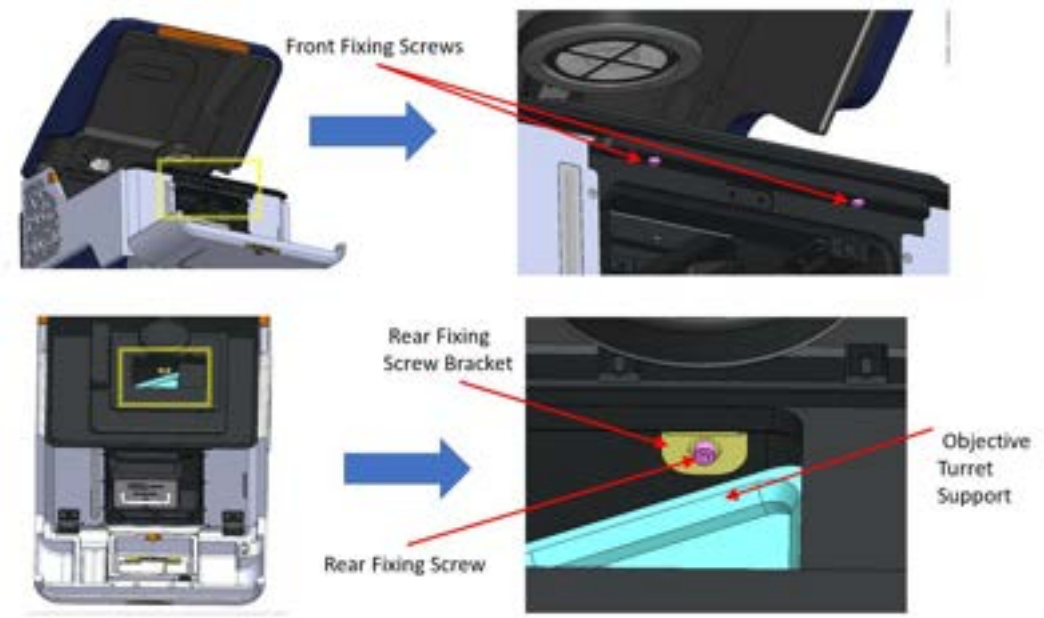

Figure 11: Schematic indicating the finger crushing hazard.

Note
If you power down the instrument then there is no hazard.

Description of Potential Hazard

If power is re-established while someone is replacing the XY Stage and a finger is placed between the objective turret support and the underside of the rear fixing screw bracket (which protrudes, although this is not immediately clear above), the Z stage could potentially home and crush a finger.

Mitigating Factors

- There is no risk during normal maintenance activity
- This is contrary to guidance in the user guide and is thus misuse
- It is a very unnatural situation to be in and is not a reasonably foreseeable scenario
- Another person would have to establish power while the first person is replacing the XY Stage, which is not a very reasonable scenario

Risk Assessment

Although possible, this is a very improbable set of circumstances and thus there is negligible risk as it is not reasonably foreseeable.

Conclusion

The hazard is such as to make reduction or elimination impractical to implement, but the risk is negligible.

1.16 Photobiological Safety

The Transmitted Light Turret has been assessed according to IEC 62471:2006 Photobiological Safety of Lamps and Lamp Systems (EN 62471:2008) and has been categorised as belonging to the Exempt Group (RG 0), "where no optical hazard is considered reasonably foreseeable, even for continuous, unrestricted use". This has relevance for the EU Artificial Optical Radiation Directive (2006/25/EC), which is an occupational safety directive in the EU, and similar regulations in other countries

1.17 Mains Safety

This product uses a standard off-the-shelf external AC/DC power supply, which is certified by multiple safety certification bodies, inc. UL, Nemko and JET. This means that as part of the manufacturer's production tests 100% of units have their insulation tested and thus no U.K. PAT testing or the equivalent is required when these are brand-new products.

This does not preclude any regular safety checks required by your national regulations.

Section 2 - Introduction

Thank you for choosing the Andor BC43 Benchtop Confocal Microscopy instrument This user guide contains useful information and advice to ensure optimum performance from your new system. BC43 integrates confocal imaging with a highly efficient dual micro lens system - pinhole disk design, outstanding sCMOS detectors and Andor´s patented Borealis uniform illumination. This combination of features makes BC43 an ideal solution for many life science imaging applications from live-cell imaging to large, thick samples. BC43 is suitable for confocal (CF) and widefield (WF) imaging or epi-fluorescence (Epi). Combined with a motorised XY stage, the instrument allows multiple point acquisition, large organism montage and multipoint scanning, resulting in outstanding productivity from a single experiment. This manual provides a description of the main features of BC43, installation, routine operation, and troubleshooting. BC43 stands for benchtop confocal, 4th generation, 3 imaging modalities.

Please feel free to contact Andor directly with any questions regarding your BC43 instrument, or via your local representative or supplier.

This manual covers the following product models: BC433.

Figure 12: BC43

Table 1: Model overview of Benchtop Microscopes Family.

Model Number	Description	Key Features	Possible Upgrades	Related Documentation
BC433	Original BC43 model	EPI fluorescence widefield Confocal Deconvolution	Super resolution	• BC43 User Guide • Fusion help
BC43 WF	Benchtop microscope with widefield	EPI fluorescence widefield	Confocal Deconvolution Super resolution	• BC43 WF,CF and SR User Guide • Fusion help
BC43 WF DC	Benchtop microscope with widefield and deconvolution	EPI fluorescence widefield Deconvolution	Confocal Super resolution	
BC43 WF SR	Benchtop microscope with widefield and super resolution	EPI fluorescence widefield Deconvolution Super resolution	Confocal	
BC43 CF	Benchtop confocal with widefield and confocal	EPI fluorescence widefield Confocal Deconvolution	Super resolution	
BC43 SR	Benchtop confocal with widefield, confocal and super resolution	EPI fluorescence widefield Confocal Deconvolution Super resolution	None	
BM42 WF	Benchtop microscope with widefield	EPI fluorescence widefield	Deconvolution Super resolution	• BM42 User Guide • Fusion help
BM42 WF DC	Benchtop microscope with widefield and deconvolution	EPI fluorescence widefield	Super resolution	

Model Number	Description	Key Features	Possible Upgrades	Related Documentation
		Deconvolution		
BM42 WF SR	Benchtop microscope with widefield and super resolution module	EPI fluorescence widefield Deconvolution Super resolution	None	

2.1 Technical Support

If you have any questions regarding the use of this equipment, please contact the representative from whom your system was purchased, or:

Europe

Oxford Instruments Andor

7 Millennium Way

Springvale Business Park

Belfast

BT12 7AL

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2.4 Trademarks and Patent Information

Andor and BC43 are trademarks of Andor Technology Ltd. Andor Technology Ltd. is a wholly owned subsidiary of Oxford Instruments plc. All other marks are property of their owners.

Manufacturers Information

Oxford Instruments Andor

Andor Technology Ltd., Belfast, BT12 7AL, UK.

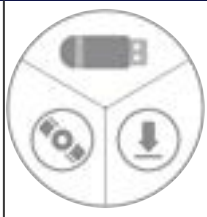
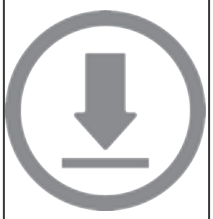
2.4.1 Patent Information

Patent Name	Country	Application No.	Published / Granted No.
Multimode fibre optically coupling a radiation source to a multifocal confocal microscope	US	US12/632,757	US8275226B2
	US	US13/603,231	US9134519B2
	CH	EP09405217.2	EP2196839B1
	DE	EP09405217.2	EP2196839B1
	FR	EP09405217.2	EP2196839B1
	GB	EP09405217.2	EP2196839B1
Imaging distal end of multimode fibre	CA	CA2779146A	CA2779146C
	GB	EP10835346.7	EP2510395B1
	IE	EP10835346.7	EP2510395B1
	DE	EP10835346.7	EP2510395B1
	FR	EP10835346.7	EP2510395B1
	US	US12/961,749	US8670178B2
	US	US14/156,742	US8922887B2
	WO	PCT/CA2010/001965	WO2011069261A1
Radiation delivery apparatus for microscope systems	GB	GB1909288.1	GB2585072B
	US	US16/911,471	US11644657B2
	DE	DE102020116802.0A	DE102020116802A1
	JP	JP2020109463A	JP2021006902A
Microscope with focusing system	GB	GB1905815.5	GB2583369B
	DE	DE102020204830.4	DE102020204830B4
	JP	JP2020076463A	JP2020197703A
	US	US16/857,218	US11500188B2
Differential Phase Contrast Microscope	GB	GB1917170.1	GB2589327B
	US	US16/953,863	US20210157114A1
	EP	EP20209921.4A	EP3828615A1
	JP	JP2020195115A	JP2021086156A

2.5 Supplied Components

The standard components supplied with BC43 are shown below. Note that BC43 is also supplied with a PC workstation.

Description		Quantity
	BC43 Model as ordered	1

Description		Quantity	Description		Quantity
	3D Navigation Joystick	1		Fusion for BC43 supplied on workstation & Imaris Quant software as ordered•2	1
	Power extension cord (3 m)	3		Microscope sample slides	Quantity depends on order
	Power supply unit for BC43	1		Universal sample holder	1
	Power distribution unit & Country specific power cord	1		User Guide (electronic copy)	1

Description		Quantity	Description		Quantity
	USB 3 cable[1] Standard-A to Standard-B cable (3 m)	1		Dust cover and lifting harness	1
	Objective lenses Always shipped with: LB CFI Plan Achro2X (LB-OBJ-02-006) Additional objectives are selectable at time of ordering.	Up to 5		Quick Start Guide	1
	Multiwell stage holder	1		Tools (Hex key, bubble level, optical swabs, T20 tool)[3]	1
	PC workstation including connection & power cables, keyboard & mouse.	1			

1. BC43 connects to the control PC using a USB 3.0 connection. This may also be referred to as USB 3.1 (Gen 1). Andor provide a USB 3.0 (Standard-A to Standard-B) cable and recommend that this is used to ensure optimum performance. Please note USB 3.0, USB 3.1 (Gen 1) and USB 3.2 Gen 1x1 are equivalent.
2. Imaris can be ordered pre installed on the PC workstation or as an option to install on a different workstation. Installation on a different workstation is described in the Imaris Welcome Letter.pdf available on the desktop of the BC43 workstation.
3. See "Tools List" on page 100

2.6 Accessories

A range of optional accessories are available to order, please contact your local sales representative for further information.

Currently Supported Objective Lenses					
Description	**NA**	**Working Distance / mm**	**Compatibility**	**Order Code**	**Supported**
2x Plan Achromat	0.06	7.5		**INS-OBJ-02-006**	NA
10x Plan Fluorite objective	0.3	16	Widefield / Confocal	**INS-OBJ-10-030**	Focus Seek & Lock
10x Plan Apochromat objective	0.45	4		**INS-OBJ-10D-045**	
20x Plan Apochromat objective	0.8	0.8		**INS-OBJ-20D-080**	
20x S Plan Fluorite objective	0.7	2.3		**INS-OBJ-20-070-LWD**	
40x Plan Fluorite objective	0.75	0.66		**INS-OBJ-40-075**	
40x Plan Apochromat objective	0.95	0.21		**INS-OBJ-40D-095**	
40x Plan Apochromat silicon oil objective	1.25	0.3		**INS-OBJ-40S-125-SIL**	Focus Lock only
40x Plan Fluorite oil immersion objective	1.3	0.24		**INS-OBJ-40-130-O**	
60x Plan Apochromat oil immersion objective	1.42	0.15	Widefield / Confocal/ Super resolution	**INS-OBJ-60D-142-O**	
100x Plan Apochromat oil immersion objective	1.45	0.13	Widefield/ Confocal/ Super resolution	**INS-OBJ-100D-145-O**	

Legacy Objective Lenses	
Description	**Order Code**
Nikon 10X 0.45NA CFI Plan Apochromat Lambda	**INS-OBJ-10-045**
Nikon 20X 0.75NA CFI Plan Apochromat Lambda	**INS-OBJ-20-075**
Nikon 60x 1.40NA Oil CFI Plan Apochromat Lambda	**INS-OBJ-60-140-O**

Legacy Objective Lenses	
Description	**Order Code**
60x Plan Apochromat objective with 0.95 NA. Working distance of 0.21 mm.	**INS-OBJ-60-095**
Consumables	
Description	**Order Code**
Immersion Oil	**INS-OBJ-OIL**
Silicone Oil	**INS-OBJ-OIL-SIL**
PC workstation data storage upgrades (add up to 1 only)	
Description	**Order Code**
4TB data storage upgrade for supplied PC work-station	**INS-PC-DRV-4TB**
8TB data storage upgrade for supplied PC work-station	**INS-PC-DRV-8TB**
Sample Holders	
Description	**Order Code**
Replacement universal sample XY-stage insert	**SV-INS-STG-UNI**
Replacement multiwell plate stage insert	**SV-INS-STG-MW2**
Incubator Sample Holders	
Description	**Order Code**
One position. 1x3 inch chamber slide holder	**MSD-INCB-1XGS-M**
One position. 35 mm Petri-dish holder	**MSD-INCB-1X35-M**
Two position. 35 mm Petri-dish holder	**MSD-INCB-2X35-M**
One position. 1x3 inch chamber slide and #2 35 mm Petri-dish holder	**MSD-INCB-GS35-M**
Open frame for multi well plates, suitable for oil immersion objectives	**MSD-INCB-MW-OIL**
Two position. 1x3 inch chamber slide holder	**MSD-INCB-2XGS-M**
One position. Lab-Tek 1x2 inch chambered cover glass holder	**MSD-INCB-1XLBTK-M**
Two position. Lab-Tek 1x2 inch chambered cover glass holder	**MSD-INCB-2XLBTK-IIM**
#1 Lab-Tek II 1x 2 inch chambered cover glass and #1 50/60 mm Petri-dish holder	**MSD-INCB-LBTK-II-60M**
#2 Lab-Tek 1x2 inch chambered cover glass holder	**MSD-INCB-2XLBTK-M**
#1 Lab-Tek II 1x2 inch chambered cover glass holder	**MSD-INCB-1XLBTK-IIM**

Incubator accessories	
Description	**Order Code**
Magnetic holder for 35 mm petri dish	**MSD-INCB-35-TL-M**
Lid with thermocouple for local / sample temperature recording at the level of the sample	**MSD-INCB-SENSOR**

Note
Objectives provided as part of your order will be calibrated for Parfocality and Parcentricity. However, to install replacement or additional objectives, an additional procedure detailed in the Fusion software user guide is required for Parfocality and Parcentration Calibration. The maximum number of objective lenses that can be installed on a BC43 system is 5.

2.7 Recommended Sample Substrates

Andor recommends and supplies the following sample substrates for optimal imaging using BC43.

Description	Order code	Manufacturer	Manufacturer order code
Rectangular Coverglasses - 22x50 mm, No 1.5 (0.16 - 0.19 mm) thick. Pack of 100.	XS-CVRSLIP-RECT	Agar Scientific	AGL462250-15
Square Coverglasses - 20x20 mm, No 1.5 (0.16 - 0.19 mm). Pack of 100.	XS-CVRSLIP-SQR	Agar Scientific	AGL46S20-15
Chambered Coverslips 8 well (Glass) Pack of 12.	XS-8WELL-CVRSLIP	Mattek	CCS-8
Chambered Coverslips 4 well (Glass) Pack of 12.	XS-4WELL-CVRSLIP	Mattek	CCS-4
Chambered Coverslips 2 well (Glass) Pack of 12.	XS-2WELL-CVRSLIP	Mattek	CCS-2
Glass Bottom Dish 35 mm. 20 mm diameter imaging area. Uncoated. Pack of 10.	XS-35MM-GLS-UTR	Mattek	P35G-1.5-20-C
Glass Bottom Dish 35 mm. 20 mm diameter imaging area. Poly D-Lysine Coated. Pack of 10.	XS-35MM-GLS-POLYD	Mattek	P35GC-1.5-14-C
96 well plate, No. 1.5 Coverslip, 5 mm Glass Diameter, Uncoated Pack of 5.	XS-96W-GLS-UTR	Mattek	P96G-1.5-5-F
24 well plate, No. 1.5 Coverslip, 13 mm Glass Diameter, Uncoated. Pack of 10.	XS-24W-GLS-UTR	Mattek	P24G-1.5-13-F
12 well plate, No. 1.5 Coverslip, 14 mm Glass Diameter, Uncoated. Pack of 10.	XS-12W-GLS-UTR	Mattek	P12G-0-14-F
6 well plate, 6-well, No. 1.5 Coverslip, 10 mm Glass Diameter, Uncoated. Pack of 10.	XS-6W-GLS-UTR	Mattek	P06G-1.5-10-F
24 well plate, No. 1.5 Coverslip, Single glass plate for maximum well imaging, uncoated. Pack of 12.	XS-24W-GLS-UTR-OIL	Greiner	662892
96 well plate, No. 1.5 Coverslip, Single glass plate for maximum well imaging, uncoated. Pack of 16.	XS-96W-GLS-UTR-OIL	Greiner	655891
96 well plate, No. 1.5 Coverslip, Single glass plate for maximum well imaging, Treated for improved cell adhesion. Pack of 16.	XS-96W-GLS-TR-OIL	Greiner	655981

Section 3 - Product Overview

This section provides an overview of the external and other features of BC43.

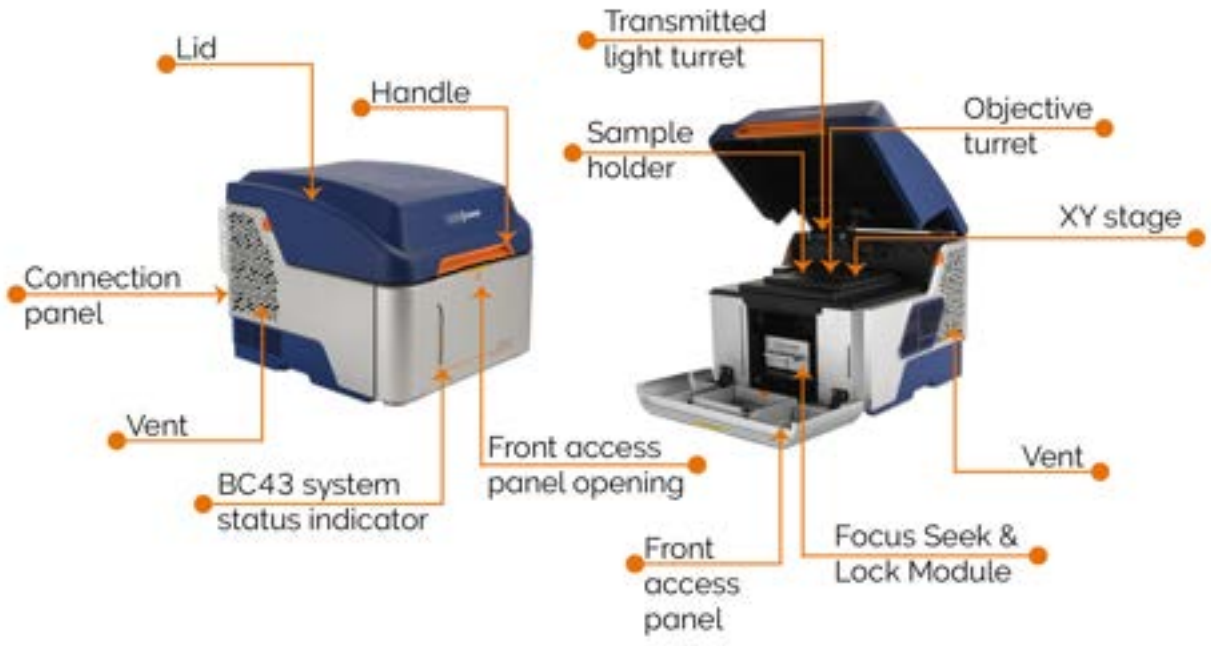


Figure 13: External features of BC43.

3.1 Power And Signal Connections

3.1.1 Connector Plate

Please see "BC43 Connections" below for an overview of the connections on the BC43 back plate.

Figure 14: BC43 Connections

Power Input

For connection to the external power supply unit (PSU) (refer to "Electrical Power Specifications" on page 221).

An On/Off switch is also present (shown above).

USB

A USB 3 compatible cable is connected between the USB socket and a PC. We strongly recommend you use the locking device built into the cable.

Please note directions for cable installation are provided in "Installation" on the next page.

Section 4 - Installation

Warning
Prior to commencing installation, the user should read the safety and warning information at the beginning of this manual.

4.1 Location

Please consider a suitable location to place BC43 in your laboratory or facility prior to unpacking the unit. BC43 and the PC workstation will require a minimum area of approximately 1.50 by 0.65 m (LxW) of desk space, but this may vary depending on the configuration. Please consider that access will be required to the rear of the unit to reach the power button. In addition to suitable space around the unit for ventilation of the equipment. BC43 can be located on a typical laboratory bench, ensure the workspace can support a ~ 70 kg instrument. For optimal image stability avoid placing directly next to other sources of vibration such as vortexes and centrifuges. When moving the instrument from outside or from a storage area that isn't close to room temperature, it is advisable to unpack the product and leave it at room temperature for 4 hours before using the instrument.

Figure 15: BC43 in place on bench

Ventilation

Ensure 250 mm clearance around unit and vents (do not block vents when operating on benchtop). Please ensure that BC43 is not placed next to or too close to the PC workstation.

Air Cooling

BC43 uses air cooling. One fan is located on the back panel of the unit to cool the laser engine. A second fan lies on the inner side of the right-hand side panel to cool the instrument's camera detector.

4.2 Unpacking BC43

Warning
BC43 requires a three-person left to get it onto the workbench.

If BC43 is being lifted or moved, at least three people are required to do this. Follow good practice for manual handling, especially the guidance provided by your employer. Ensure that BC43 is placed on a level surface.

BC43 is shipped with a lifting harness and dust cover. To remove BC43 from the shipping packaging please use the supplied lifting harness straps. One set of lifting handles are provided per person and a three-person lift is required, so 6 harness straps in total. Each person lifting the unit should lift a pair of straps of the same colour: either blue, green or black.

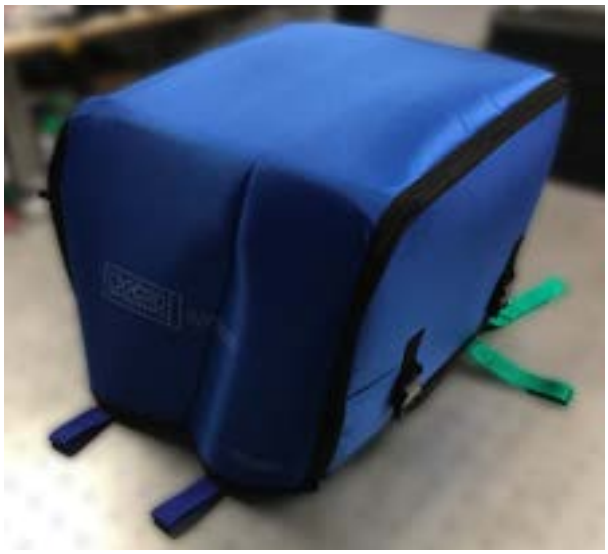


Figure 16: BC43 unit on optical table, with dust cover and two out of three pairs of lifting harness straps visible (blue and green).

Warning
The dust cover is attached to the lifting harness and must not be removed prior to lifting and placing BC43 on the workbench on which it is to be used.

Once BC43 has been placed on the bench and moved into position the top part of the dust jacket can be unclipped from the base and then unzipped.

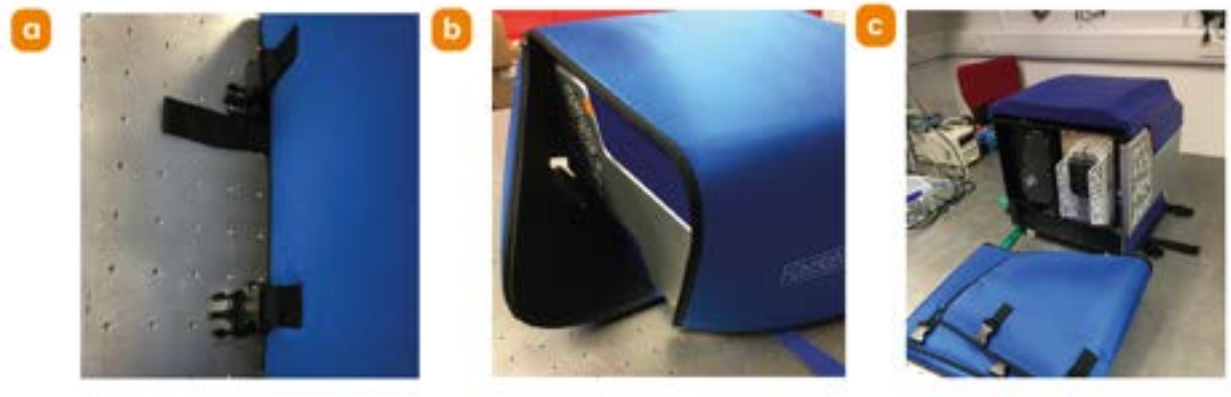


Figure 17: Removing the dust jacket on BC43. a) Unclip the side straps. b) Unzip the side panel. c) Remove the dust jacket.

The base can be slid out from under BC43.

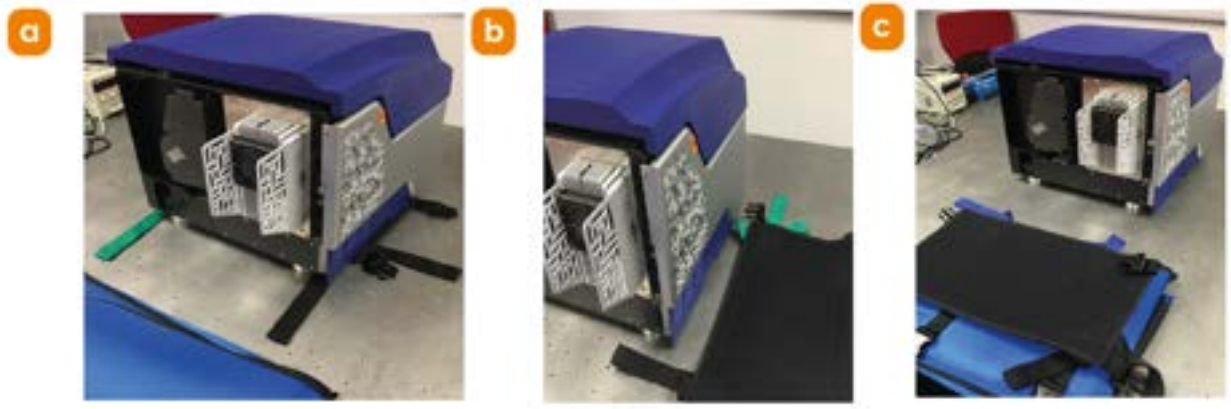


Figure 18: Remove the base of the lifting harness. a) and b) Slide out the supporting base panel. c) Store the base with the dust jacket.

If required BC43 can be levelled using the adjustable feet, see "a) Bubble level. b) Adjustable foot positions. c) Foot adjustment mechanism." on the next page Open the top cover lid and place a small bubble spirit level (provided) on the XY stage. If the bubble is not in centred in the level then, insert a Small Flat Head Screwdriver into the indicated through holes of the front feet and sequentially turn both feet clockwise or anti-clockwise until the spirit level indicates the XY stage is level to the workbench/desktop.

Figure 19: a) Bubble level. b) Adjustable foot positions. c) Foot adjustment mechanism.

4.3 Assembly

4.3.1 BC43 and PC Workstation Setup

Please see the supplied quick start guide for set up instructions for BC43, available online at: andor.oxinst.com/downloads

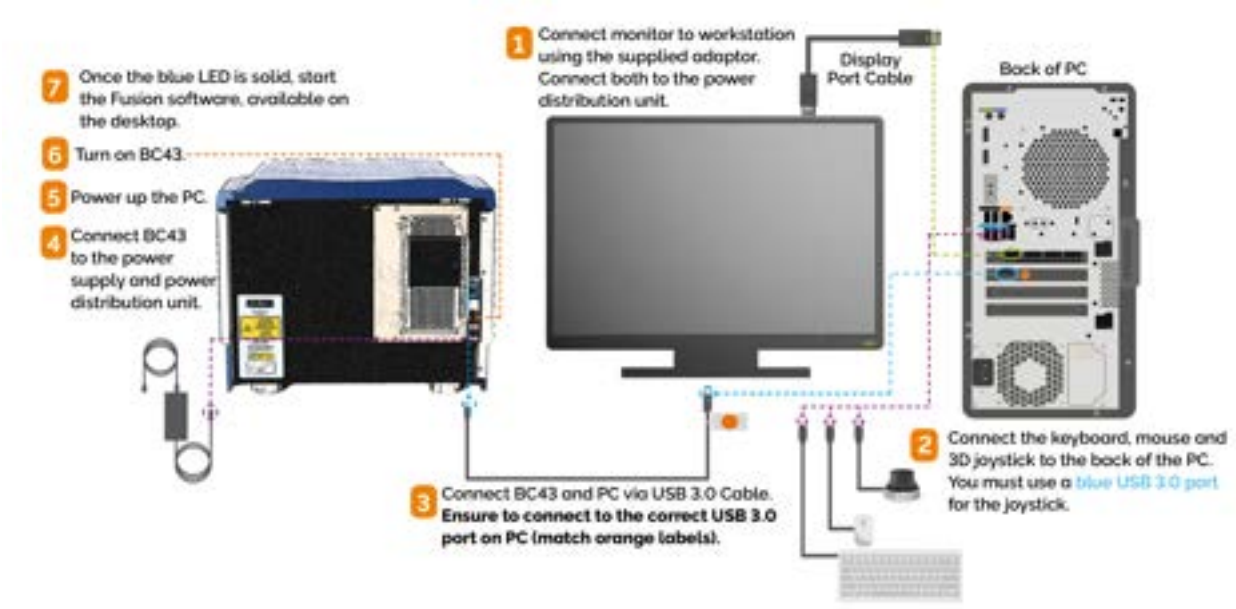


Figure 20: Connections between PC workstation (HPZ2G9), BC43, accessories and power supplies. See "Connection Diagrams for Previous PC Models" on page 224.

4.3.2 Connect and Configure the 3D Connexion Spacemouse

The 3D Connexion Spacemouse is already installed and configured with BC43, the provided PC workstation and Fusion. See "Fusion Software" on page 62 in the default Windows login account of the PC workstation provided with the BC43. Please note that the 3D Connexion Spacemouse joystick MUST plug into a USB 3.0 port on the PC workstation. Please check the connection diagram carefully in "BC43 and PC Workstation Setup" on the previous page. Insertion of the of the joystick USB connector into a USB 2.0 port will result in irregular XY stage movement! If required in the future please see the 3D Connexion website for further information on drivers: https://3dconnexion.com/uk/drivers/

Figure 21: Spacemouse, left-hand button toggles XY – Z, and right-hand button toggles sensitivity.

Note
Creation of a new Windows login account on the PC workstation (See ""Creating Additional Windows Accounts" on page 61") requires that the 3D Connexion Spacemouse be properly configured to operate in Fusion.

Follow the instructions below to assign the left joystick button to toggle the motion between the XY stage and Z focus motor, and to assign the right joystick button to control the XY/Z motion sensitivity (course/fine/super-fine).

1. Ensure Fusion is open and running on the PC workstation.
2. Search for "3Dconnexion" in the Windows search bar.
3. Open the 3Dconnexion app.

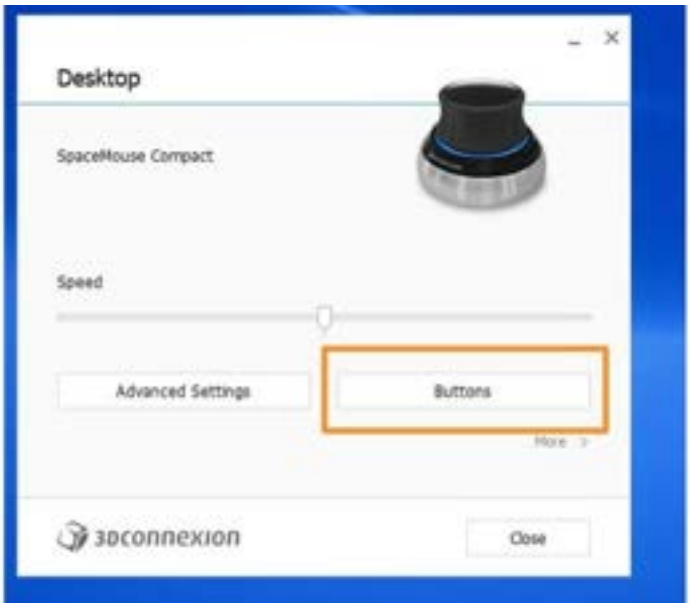


Figure 22: In the 3Dconnexion app window select "Buttons".

4. Maximise then minimise the Fusion interface in Windows.

Figure 23: Check the Fusion logo appears as shown in image. If it does not, check that Fusion is running and relaunch the 3Dconnexion app.

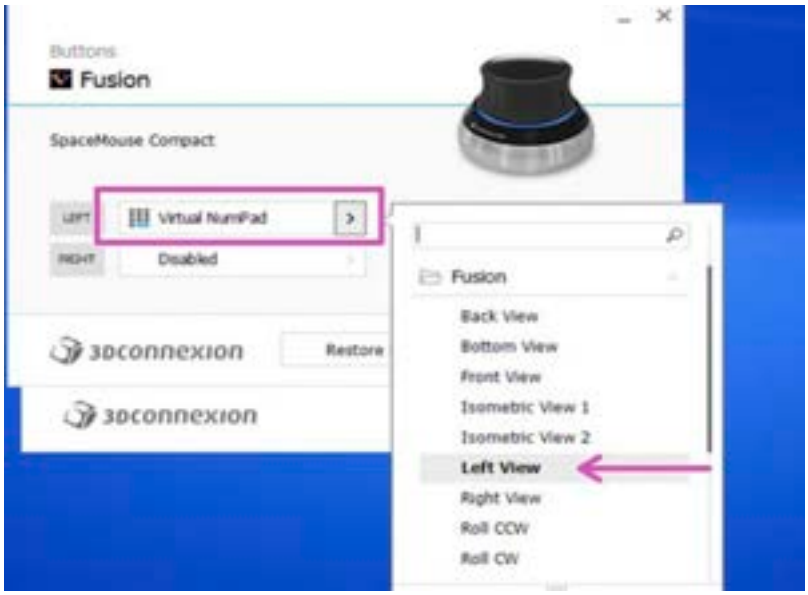


Figure 24: From the left drop-down list select 'Fusion' and 'Left View'.

5. From the right drop-down list select 'Fusion' and 'Right View'.

Figure 25: Confirm that both options are set as shown in the image.

6. Verify that the 3Dconnexion buttons now work as expected with the Fusion interface.
7. Close the 3Dconnexion app.

4.4 PC Workstation Operating System Notes and IT / Networking

4.4.1 Windows Updates

The BC43 instrument control (Fusion) and visualization/analysis (Imaris) software run in the Windows™ operating system environment of the PC Workstation. Periodically, Microsoft will release operating system feature or security updates which automatically download onto the PC workstation.

To prevent interruption of critical long-term imaging experiments, it is recommended to check for Windows updates and immediately install BEFORE starting the experiment, or manually set an update installation delay time that exceeds the expected duration of the experiment. Please also see Regular Checks "Windows Operating System Updates" on page 140.

Check for Windows updates by first clicking on the Start Menu icon and selecting the Settings option:

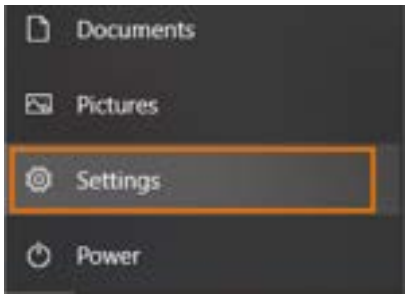


Figure 26: Start Menu icon, with the Settings option selected.

In the Windows Settings, click the Update & Security option:

Figure 27: Update & Security option

In the Windows Update menu, click the Check for Updates button to scan for and download current updates.

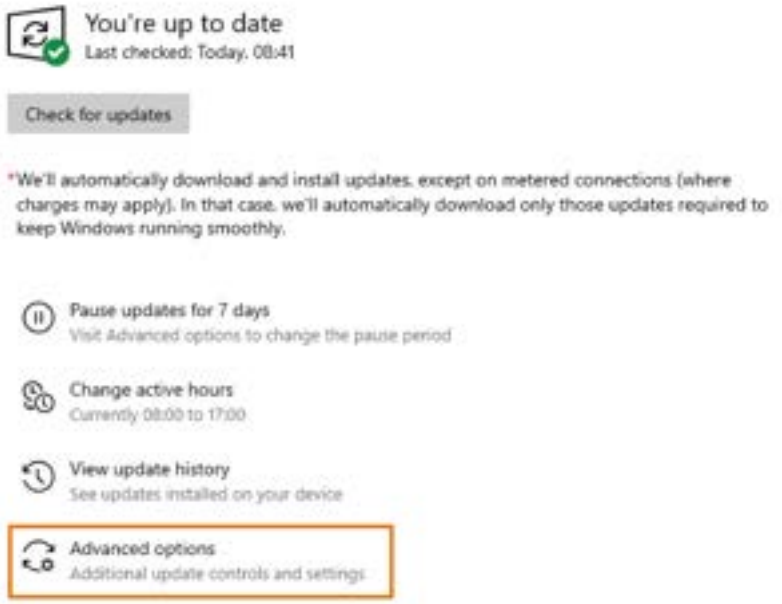


Figure 28: Check for updates and Advanced options highlighted.

If any updates are downloaded, they can be installed immediately followed by a restart of the PC workstation, or alternatively a delay can be set for the installation after the experiment by first clicking on Advanced Options in this menu, and then selecting an install date under the drop-down menu under the Pause Updates heading. The installation of Windows updates can be delayed up to 35 days.

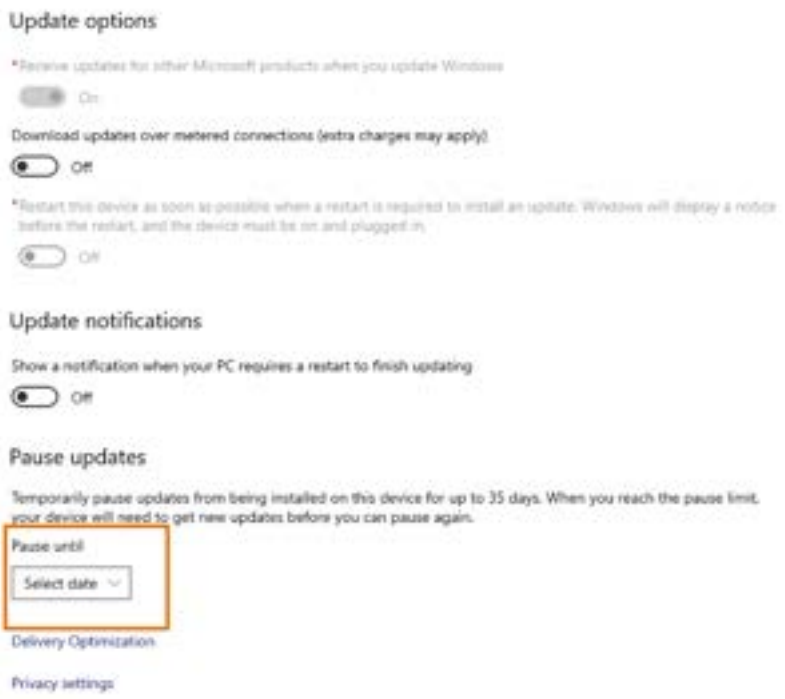


Figure 29: Advanced options, with Pause updates until options highlighted.

Should a new Windows operating system become available, please contact your local Andor service provider to avoid any software compatibility issues.

Windows support for both Fusion and Imaris is documented on the product websites:

fusion-benchtop-software-guide.scrollhelp.site/fusionum/system-requirements

imaris.oxinst.com/support/system-requirements

4.4.2 Anti-virus software

It is common for facility IT/network administrators to require the addition of anti-virus software onto all site computer workstations. However, anti-virus software has the potential to disrupt the normal operation of the image acquisition software for the microscope (Fusion) or the image analysis software (Imaris). By default, Fusion and Imaris are added to the allowlist of Windows Defender, the native anti-virus program for the Windows operating system. It is highly recommended that Fusion and Imaris be added by the IT/network administrators to the allowlist of any site-specific or mandatory anti-virus applications and that virus scans should be scheduled outside of working hours on the BC43 PC workstation. It is also suggested that the anti-virus software be disabled for any critical extended or overnight imaging sessions.

4.4.3 Creating Additional Windows Accounts

The BC43 image acquisition software ("Fusion Software" on the next page) uses the Windows login to determine the current user. Creation of new Windows user accounts depends on how the local IT/networking administrator plans to configure the system onto a domain network or not. Please refer to the online Fusion for BC43 software guide for proper multi-user Windows accounts setup: fusion-benchtop-software-guide.scrollhelp.site/bc43softwareguide/multi-user

Warning
Failure to follow the proper multi-user setup instructions may result in software failure or inaccessibility of the software to new users and create unnecessary system downtime.

4.4.4 Changing the PC Workstation Name or Media Access Control (MAC) Address

The image acquisition software (Fusion – see "Fusion Software" on the next page) licence key is node-locked to the PC workstation. This type of software license ensures the software will run on only one specific host computer. The license key is tied to the computer's Host ID and is stored on the computer's hard disk. Therefore changing the PC name or its MAC address may break the licence. It is highly recommended the local IT/networking administrator review the Fusion for BC43 software guide's licensing information: fusion-benchtop-software-guide.scrollhelp.site/fusionum/licensing

Warning
A new license must be issued to the PC workstation if the computer or MAC address is changed. The software will be inaccessible until a new license key is properly registered.

4.5 Fusion Software

Fusion is Andor's image acquisition and control software for microscopy instruments and comes pre-installed on the supplied PC workstation. Before starting Fusion, ensure that the blue LED, at the front of the BC43, is solid. See the connection diagram"Assembly" on page 54 for full guidance. Please see the following sections to get started using Fusion with BC43. Note that this is a brief introduction to the Fusion user interface and presents essential concepts to get started using your BC43 instrument. Please see the full Fusion documentation available at fusion.help.andor.com for more information on Fusion. For Fusion software features specific to BC43 please see fusion-benchtop-software-guide.scrollhelp.site/bc43softwareguide/

4.5.1 Fusion User Interface

To open Fusion double click on the software icon on the supplied PC workstation desktop.

Figure 30: Fusion short cut on supplied PC workstation desktop.

The main working areas of the software are shown below. Please note the expander panel can be hidden or expanded as required using the arrow on the right-hand side of the Fusion screen.

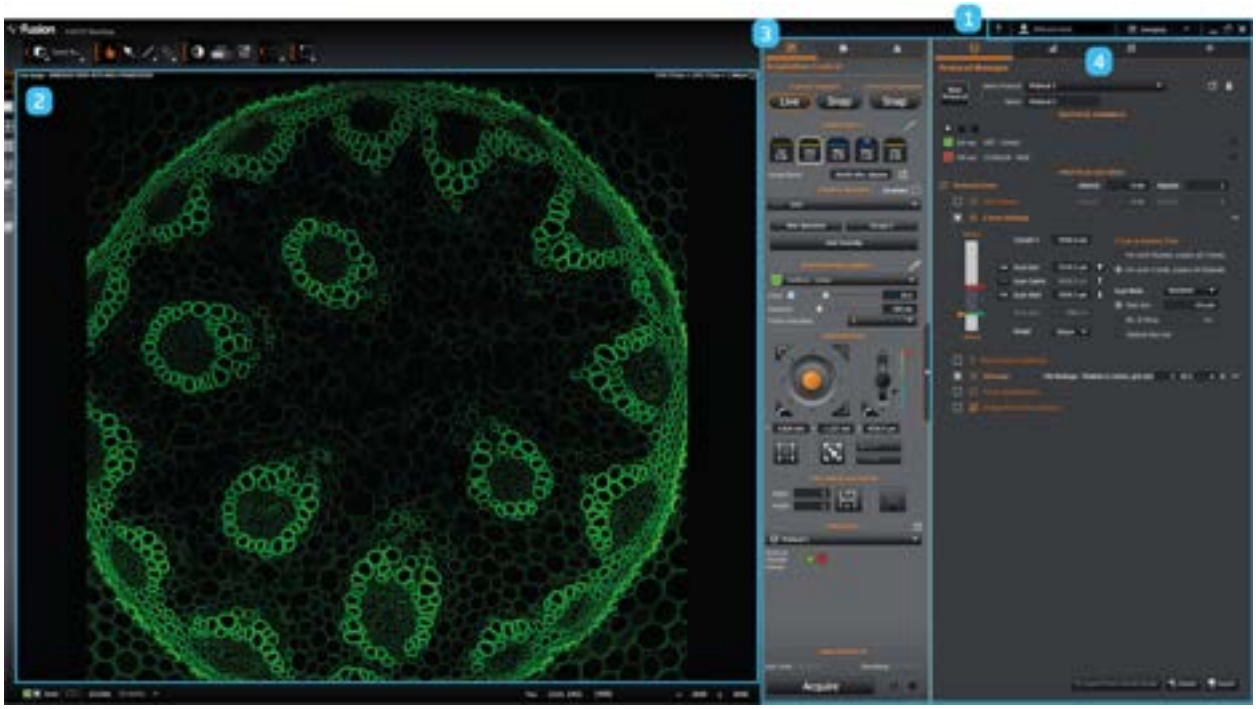


Figure 31: Fusion interface: 1. Application bar, 2. Image viewer panel. 3. Remote control acquisition panel. 4. Expander panel.

4.5.2 Application Bar

From the application bar ("Application bar screen shot. Access the 1. Fusion documentation and 2. the preferences drop down." below) the user can access the Fusion user documentation (question mark icon) and the preferences tab (wheel icon).

Figure 32: Application bar screen shot. Access the 1. Fusion documentation and 2. the preferences drop down.

4.5.2.1 Preferences Tab

The preferences tab can used to define general image acquisition settings such as where images are saved on the computer hard drive. To access the preferences tab, select the wheel from the application bar and the preferences menu will fill the screen.

In display settings User Interface Zoom can be used to adjust the size of the Fusion interface on the PC workstation monitor. Adjust this display setting to optimise the user experience.

Within File Manager, select the desired Root Folder to save acquired images. Fusion can automatically create subfolders for image storage as selected by the user. For example, a subfolder could be created on a daily, weekly or monthly basis. Please note this feature can be turned off by selecting none from the dropdown.

Next, decide what should be added to the image name. For example, Channel, Date, Protocol, Time or User. Multiple selections can be made, and an example of the naming convention will be shown below depending on the selection. If no image identifier options are selected, the image name will be automatically updated with sequential numbers during the acquisition process. Click the Back to Imaging button when all preference settings have been finalized.

For more information on Preferences, please see the fusion user manual available at the following link: fusion-benchtop-software-guide.scrollhelp.site/fusionum/preferences

4.5.3 Image Viewer Panel

The image viewer panel allows the user to view the current field of view of the BC43 instrument and also to display images that have already been acquired.

For more information on specific functionality of the image viewer panel, please see the Imaging section of the fusion user manual available at the following link: fusion-benchtop-software-guide.scrollhelp.site/fusionum/imaging

4.5.4 Remote Panel

Please use the Remote Panel to access Navigation and Imaging controls, File Manager, and Notifications, as shown below.

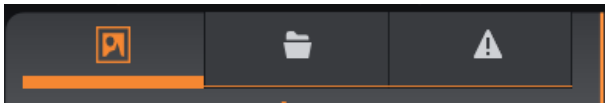


Figure 33: Remote control acquisition panel, tabs. From Left to Right: Acquisition control, File Manager, Problem Notifications.

For more information on specific functionality, please see the Remote Panel of the fusion user manual available at the following link: fusion-benchtop-software-guide.scrollhelp.site/fusionum/remote-panel

4.5.5 Expander Panel

At the top of the Expander Panel the user can access the Protocol Manager tab, the Analysis tab and the Image Processing tab.

Figure 34: Expander panel, Left: closed. Right: open.

For more information on specific functionality, please see the Expander Panel section of the fusion user manual available at the following link: fusion-benchtop-software-guide.scrollhelp.site/fusionum/expander-panel

4.6 Add Objective Lenses to the Microscope

The objective lenses are shipped separately from the main unit during transit and must be self-installed. The ordered microscope objective lenses will be preprogrammed in Fusion and available to select in the Remote control acquisition panel. Please use the following instructions to add the objective lenses to the BC43 unit in their correct positions.

1. Ensure the PC is powered On.
2. Turn on BC43 using the switch at the back of the unit.
3. Once the blue LED light is continuously illuminated in blue the BC43 unit is ready to be connected to the software.
4. Start Fusion, using the short cut installed on the desktop of the supplied workstation.
5. Open the BC43 unit.
6. Remove the sample holder, see Figure below.

Figure 35: a) Grab hold of the front edge of the sample holder with two hands. b) Pull up and away to release the sample holder from the sample stage. c) Fully remove and set aside the sample holder.

Warning
IF THE SAMPLE HOLDER IS NOT REMOVED THE OBJECTIVES MIGHT BECOME DAMAGED DURING THE INSTALLATION PROCEDURE.

7. Within Fusion select the air (dry) immersion objective to be installed. Clicking the selected objective icon will rotate the objective turret in the BC43 to the correct engaged position corresponding to the selected objective lens.

Figure 36: Objective selection in Fusion

8. Screw the objective lens clockwise onto the engaged turret position until it catches on the brass screw threading. Continue to turn the objective lens barrel clockwise until it can turn no more and is secured in place.

Figure 37: a) Empty objective turret with nosepiece cap. b) Empty objective turret with nosepiece caps removed. c) Turn the objective clockwise to install.

9. Repeat steps 7 - 9 for all air (dry) immersion objective lenses.

10. If present, select the objective which uses an immersion media. Fusion will flag a prompt, see Figure below. Select Skip & Continue to avoid moving the hardware to the add oil position which would make installation of the objective more difficult.

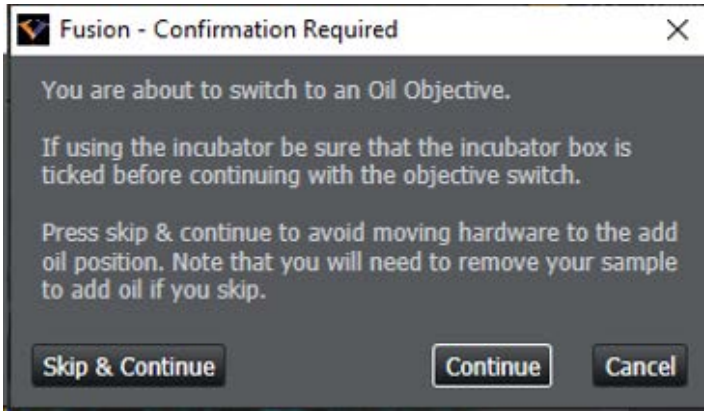


Figure 38: Fusion prompt for oil objective.

11. Install the immersion media objective.
12. Ensure that the objective lenses are placed in the correct location, each position in Fusion and objective lens are numbered.

Warning
BC43 IS AN AUTOMATIC MICROSCOPE, AND IN ORDER TO AVOID POTENTIAL OBJECTIVE DAMAGE THE OBJECTIVES MUST BE PLACED AT THEIR SPECIFIED LOCATION ON THE MICROSCOPE TURRET.

4.7 Add a Sample

BC43 is supplied with a standard and multi-well sample holder. The standard insert can hold traditional 1" x 3" glass microscope slides and coverslip-bottomed culture dishes. The multi-well sample holder is designed to support traditional 127.15 mm x 85.05 mm microplates. Use the recommended sample substrates, "Recommended Sample Substrates" on page 46, for optimal imaging results. Please use the following set of instructions to place a sample slide within BC43.

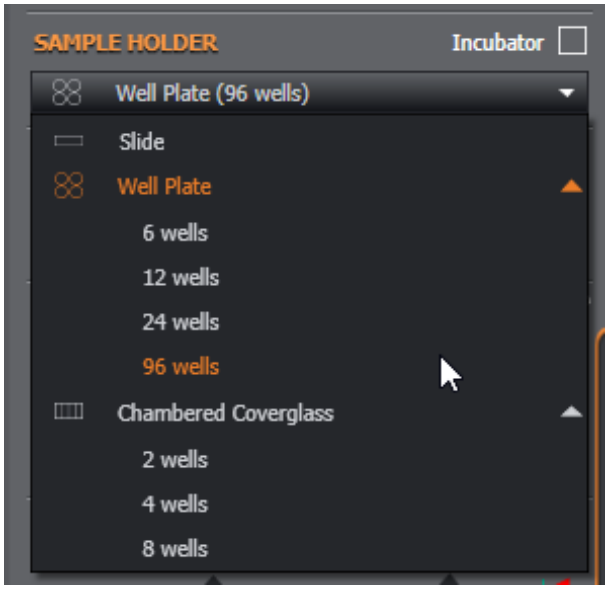


Figure 39: Sample holder options in Fusion for BC43.

1. First select a sample from the provided microscope slides.
2. Select the 2x Objective within Fusion.
3. Open the BC43 lid.
4. Place the sample in the sample holder, (note, there is no need to remove the sample holder).
5. Ensure that you place the sample with the coverslip facing down towards the objective lens.

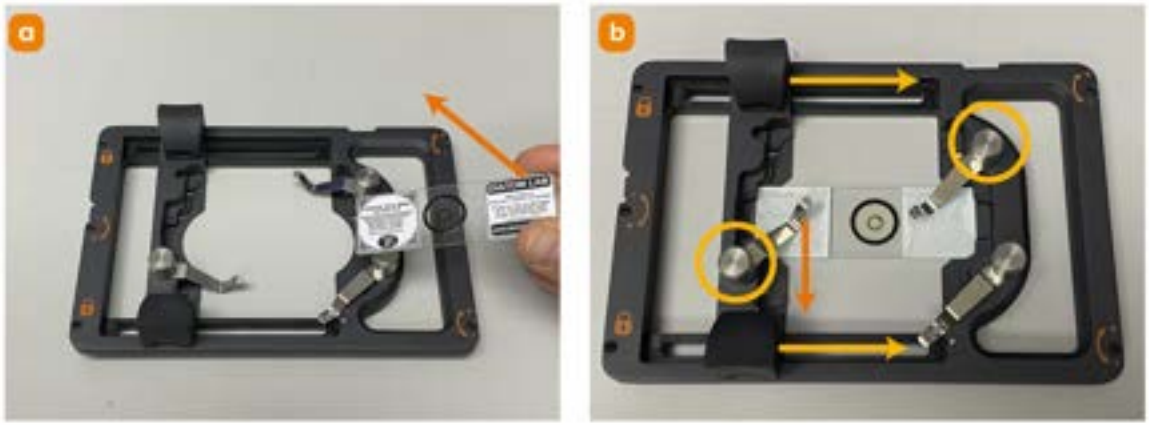


Figure 40: Insert the sample in the sample holder. a) ensure to insert the sample with the coverslip facing down. b) Slide the left hand side of the sample holder to adjust for the size of the sample slide. Slide the clips to secure the sample in the sample holder.

Note
The right-hand side of the sample holder is fixed and the left-hand side can be adjusted to accommodate the size of the sample slide.

6. Use the aluminium clips to secure the sample to the sample holder.
7. With the lid of the BC43 open, use the 3D joystick to manoeuvre the sample over the objective. Look into the BC43 unit to see when your sample is over the objective.
8. Close the BC43 lid and see the next section.

For further guidance on using the sample holder please see our Sample Holder and Joystick Video Tutorial.

4.8 Align the Sample for Imaging

To add your sample to BC43 please follow the directions in "Add a Sample" on page 71 prior to starting these alignment procedures.

4.8.1 Alignment Using the Fusion Interface

1. Within Fusion, ensure the 2x objective lens is selected.
2. Using the joystick – roughly place the sample aligned with the 2x objective.
3. Close the BC43 lid.
4. Select the appropriate channel - Press live, to find the sample select DPC or brightfield (the transmitted light channels).

Figure 41: Navigation channel settings in Fusion.

5. Use the navigation tool to move in XY and centre the sample. Navigate in the XY plane by dragging the orange highlighted joystick or using controls as detailed below.

Figure 42: XY navigation toggle in Fusion highlighted in orange. On the blue square, click to modify XY step size (S= super-fine, F=fine, C=coarse). Alternatively click on outer arrows in purple circle to move in large steps of 10 µm. Click inside area of amber circle to finely adjust position by 0.1 µm (button will highlight as cursor hovers over it).

6. If required, focus the sample in the Z plane. To focus the sample, click or drag on the orange circle in the middle for Z movement of the objective through the sample or use the controls as shown below.

Figure 43: Z Navigation toggle in Fusion highlighted in orange. On the blue square, click to modify Z step size (S= super-fine, F=fine, C=coarse). Alternatively, click on the arrows to move to the desired position. Click on the black arrow in the orange box to move in larger steps. Click on the grey arrow in the purple box to move in smaller steps (10x less than external arrow).

7. Click on the  icon (in the specimen overview section) to create an overview of the sample.
8. Adjust the sample focus, then switch to the 10x objective lens, and proceed to image acquisition.

4.8.2 Alignment Using the 3D Joystick

1. Within Fusion, ensure the 2x objective lens is selected.
2. Ensure that the joystick is placed on the proper orientation (i.e. the connecting wire is pointing away from the user).
3. The BC43 joystick has 2 controllers activated:

   Left-hand side click - toggles navigation (or Focus) speed (S= super fine, F=fine, C=coarse)

   Right-hand side click – toggles between XY and Z movement.
4. Position the sample over the 2x objective lens by clicking and activating the appropriate controls on the joystick.
5. Close the lid of the BC43.

Note
The user can align the sample with the lid of the instrument closed.

6. Select an appropriate imaging channel to view the specimen, then press the Live button at the top of the Fusion Remote Panel.

Figure 44: Navigation channel settings in Fusion

7. If the sample is already in place the user can toggle to focus.
8. If the sample is not in place, on specimen overview (under the navigation tab) click on the icon  to create a low-resolution overview of the sample.
9. Select the click to navigate mode  using this button then click on the centre of the sample to navigate the stage to this position.

10. Adjust the sample focus, then switch to the 10x objective, and proceed to image acquisition.

4.8.3 Description of the Focus Seek and Lock (FSL) Module

The Focus Seek and Lock (FSL) module, located directly below the objective lens turret carousel ("External features of BC43." on page 47), operates by directing near infrared (NIR) light of a light-emitting diode (LED) towards the sample where it is back-reflected from the sample. This back-reflected NIR light is monitored by the system detector, and tracks the lowest coverslip surface relative to the objective lens. The FSL module exploits this in two ways: The Find Coverslip feature and the Focus Stabilization feature.

When the Find Coverslip command is activated, the FSL module determines the Z-motor position that corresponds the bottom of the coverslip position as described above, and then the focus drive is offset upwards by a pre-determined offset corresponding to the thickness of the coverslip to bring the bottom of the sample into focus. Note: The Find Coverslip command is not compatible with the 2x and immersion oil objective lenses.

When the Focus Stabilization option is activated within an imaging protocol, at the beginning of the protocol the FSL module will determine the Z-motor position which corresponds to the bottom of the coverslip and references it relative to the focus position set by the user. Every 10 minutes, the Focus Stabilization routine pauses the protocol for a short period (~20-30 seconds) and re-checks the position of the bottom of the coverslip. If the coverslip position is unchanged, no action is taken and the protocol resumes. Conversely, if the coverslip position has altered due to Z-motor thermal drift, the objective lens focus is automatically adjusted by moving the Z-motor to match the coverslip-focus lock offset distance once again. Note: Focus Stabilization is only active during imaging protocols and applied to the current Z-position or the Z-positions associated with the Multi-Position Settings option of the protocol manager (if activated); The focus is not locked during a live preview of the specimen.

Please refer to the online Fusion for BC43 manual for further details:

fusion-benchtop-software-guide.scrollhelp.site/bc43softwareguide/focus-stabilisation

Plastic-bottom sample substrates are compatible with the FSL only when using dry (non-immersion) objective lenses. When using the Find Coverslip feature with a plastic-bottom substrate, the FSL will focus the instrument to the bottom of the substate, but the user must adjust the focus to the plane of interest within the sample if the thickness of the plastic bottom exceeds 170 µm. Best imaging performance results when using #1.5 (0.16 - 0.19 mm thick) glass coverslip based

substrates. Use the recommended sample substrates in the table in "Recommended Sample Substrates" on page 46, for best performance.

4.8.4 Set up Sample Navigation Bounds

After collection of a montage or when working with a big sample area, to constrain the navigation area to the most relevant section of the sample please select the square button  from the Navigation panel. Once selected the button will become highlighted in orange and a white, rectangular box will appear in the Image viewer panel. Please move this to include the relevant area of the sample, over which navigation will be restricted to. Once selected press 'Enter', the bounds will be set and the region selected will now be surrounded by an orange container.

To modify the bounds please use the '**Unlock**' button, and repeat the procedure detailed above. To clear the bounds press the '**Clear**' button.

4.9 Create a New Protocol

BC43 has two fluorescence imaging modes: confocal (CF) and widefield imaging or epi-fluorescence (Epi). For each imaging mode the user can acquire 4 different channels,

CF	Indicates the selection of a channel in confocal mode
Epi	Indicates the selection of a channel in widefield mode

The laser lines and emission filters corresponding to the specific channels are:

Channels	Laser Lines / nm	Emission filters / nm	Typical Imaging Fluorophores
Confocal -Blue/ EPI - Blue	405	445/20	DAPI
Confocal - Green/ EPI - Green	488	529/24	GFP, Alexa Fluor 888
Confocal - Orange/ EPI - Orange	561	595/31	TRITC, Texas Red, mCHERRY, Alexa Fluor 568, 594
Confocal - Red/ EPI - Red	638	708/75	Alexa Fluor 633, 647

These are representative fluorophores, but operation of the system is not restricted solely to them. Consult with your application specialist to discuss further options.

The user also has the option to combine confocal or widefield imaging with a transmitted light technique. BC43 has two different transmitted imaging modalities: Brightfield or DPC (Differential Phase Contrast).

1. Open the Expander Panel, please select the New Protocol button, see "Protocol manager settings, New Protocol button highlighted." below

Figure 45: Protocol manager settings, New Protocol button highlighted.

2. Click on the + icon to add the channels required for the acquisition, and if desired use the arrows to rearrange the order of acquisition, see "Protocol manager settings, channel additional and acquisition order buttons

highlighted." on the next page

Figure 46: Protocol manager settings, channel additional and acquisition order buttons highlighted.

3. Choose the type of protocol needed, tick on the appropriate box.
    a. More than one type can be selected, and the user can combine:
        i. Montage with Z-scan,
        ii. Z-scan with time lapse,
        iii. Z-scan with time lapse, and montage
    b. Click on time series and set the time interval and number of repetitions
4. To acquire a time-series protocol.

Figure 47: Protocol manager time-series settings.

5. To acquire a Z-scan protocol

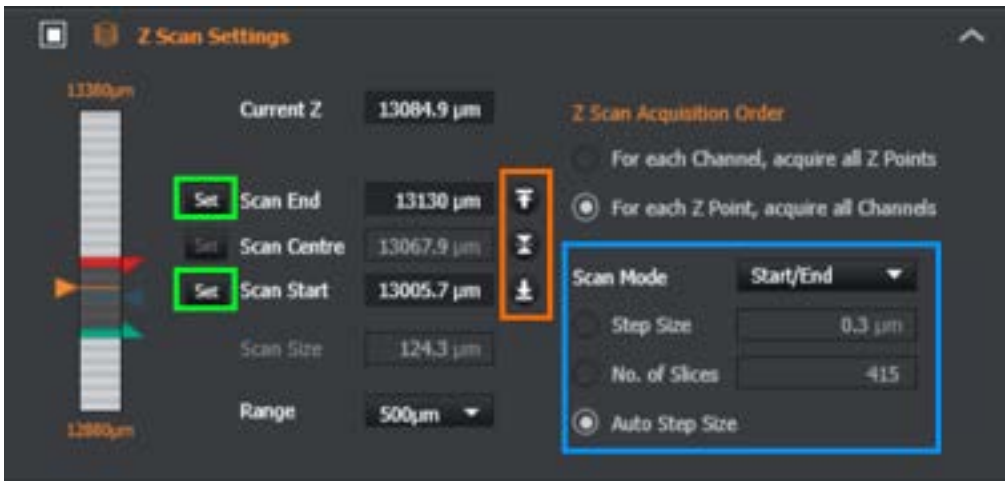


Figure 48: Z scan settings.

6. To acquire a multi-position protocol for a slide:

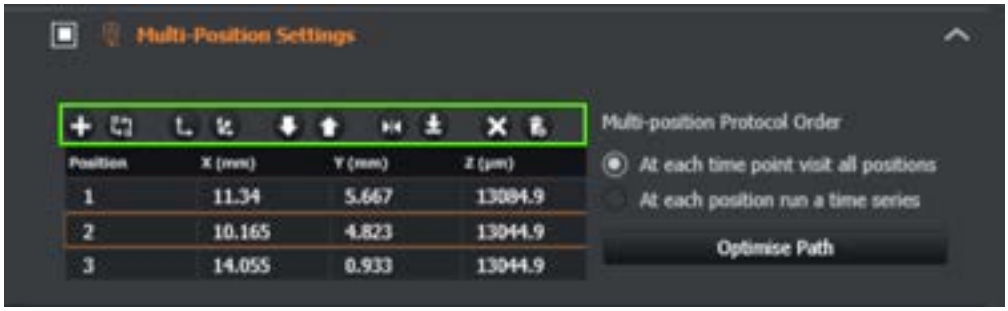


Figure 49: Multi-field position settings in Fusion.

7. To acquire a stitched montage protocol:

Figure 50: Montage settings in Fusion. Right perimeter settings.

Figure 51: Image processing settings in Fusion.

8. For time series and montage, it is useful to activate Focus stabilization, "Focus stabilisation options." below

Figure 52: Focus stabilisation options.

9. On the remote panel, create an image name to be associated with the result of the protocol acquisition:

Figure 53: Image name settings.

10. To Run the protocol press acquire, "Acquire button." below

Figure 54: Acquire button.

Please see the Fusion user guide for further information on setting up protocols fusion-benchtop-software-guide.scrollhelp.site/fusionum/creating-a-protocol

4.10 Quick Acquisition

On the remote panel, a quick acquisition can be performed (see "Quick acquisition on the remote panel." below).

1. Press Snap – a single image of the current field of view will be acquired with the selected Navigation channel.
2. Press protocol channels, a snap image of all the channels in the selected protocol will be acquired.

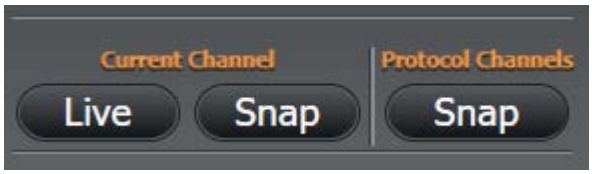


Figure 55: Quick acquisition on the remote panel.

More information at: fusion-benchtop-software-guide.scrollhelp.site/fusionum/snapping-an-image

4.11 View Acquired Images

To view acquired images click the File Manager tab (folder icon) at the top of the Remote Control Acquisition Panel, see "Viewing acquired images options under the file manager tab." below. From here, select which images to keep for further analysis. Images can be opened, in the Image Viewer Panel, via an exported file or opened directly in Imaris.

Figure 56: Viewing acquired images options under the file manager tab.

4.12 Supported File Export Formats

Image File Name	File Extension
Imaris	.IMS
Tagged Image File Format	.TIFF

More information at: fusion-benchtop-software-guide.scrollhelp.site/fusionum/supported-file-formats

4.13 Accessories Installation

4.13.1 Adding the Okolab Incubator

Read before starting procedure	
When should the procedure be performed?	Install and use the incubator when temperature control of the sample environment is required. The incubator is an optional accessory and not supplied as standard please see "Accessories" on page 43. Ensure to install on top of the objective two hours prior to intended use, to guarantee temperature stabilisation. To access the latest manual for the Okolab incubator please visit the Okolab support page. The following procedure outlines how to install the incubator with an objective heater but without gas or CO_2.
Operation difficulty	Moderate
Time to complete	20-30 mins
Tools	None
Laser safety considerations	**Class 2 laser radiation is accessible with the lid open at the microscope objective lens or objective lens port, but only one laser can be operational, see "Operational laser access. a) Sample holder and b) Focus Seek and Lock opening." on page 17.**
Mechanical safety considerations	None

1. The unit can be powered on for this procedure.
2. First completely remove the sample holder from the XY stage.
3. Gently push the incubator chamber into the XY stage opening until it clicks into place.

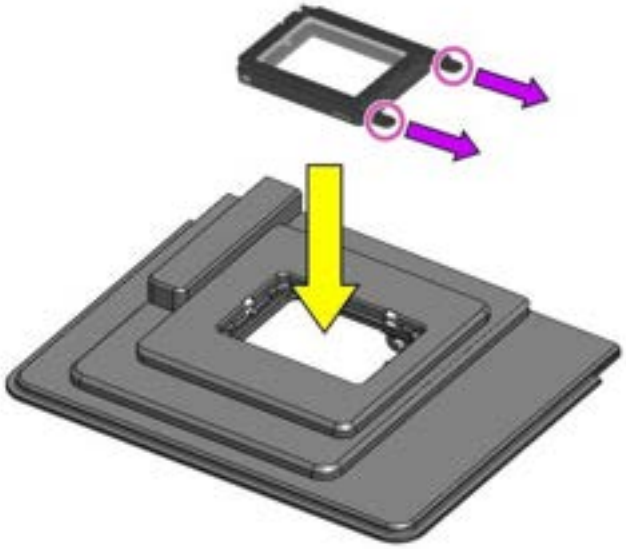


Figure 57: With the incubator oriented correctly, gently push the device into XY stage opening until it clicks into place.

4. Cable management. First run the cables and wires from the stage-top incubator into the groove at the back end of the transmitted light LED turret. Provide some slack in the cables to allow the stagetop incubator to travel with the full range of the XY stage. Please see "Thread the wiring of the incubator over the back of the BC43 instrument." below Next, feed the cables through the "W"-shaped groove at the top of the rear chassis all the way over to the orange incubator access ear. Finally, pass the cables through the incubator access ear hole, this is so the lid can be closed without trapping the cables.

Figure 58: Thread the wiring of the incubator over the back of the BC43 instrument.

5. Locate and remove objective heater cable holster. To install the objective heater follow the procedure in "Replacing the Objective Heater Holster" on

page 150.

6. Objective heater cable front panel routing. Please note the following instructions describe how to feed the cables of the immersion objective lens heater into the BC43. These steps can be ignored if using a dry/air objective lens.

Figure 59: a) Route the objective heater cable through the lower-left corner of the BC43 unit and feed the cable through the grooves in the backside of the front panel access door. b) Bring the cable to the middle of the backside of the door and pass it under the orange swing-out latch. Close the latch once the cable is through.

7. Load terminal end of objective heater cable into holster, as shown in "a) Fit the terminal end of the objective heater cable connector into the objective heater cable holster. b) The orange cable leading to the wrap-around heater should hang out of the shaft end of the holster. Now reinsert the holster back into the front chassis of the BC43." on the next page Full instructions for removal/insertion of this part can be found in "Replacing the Objective Heater Holster" on page 150.

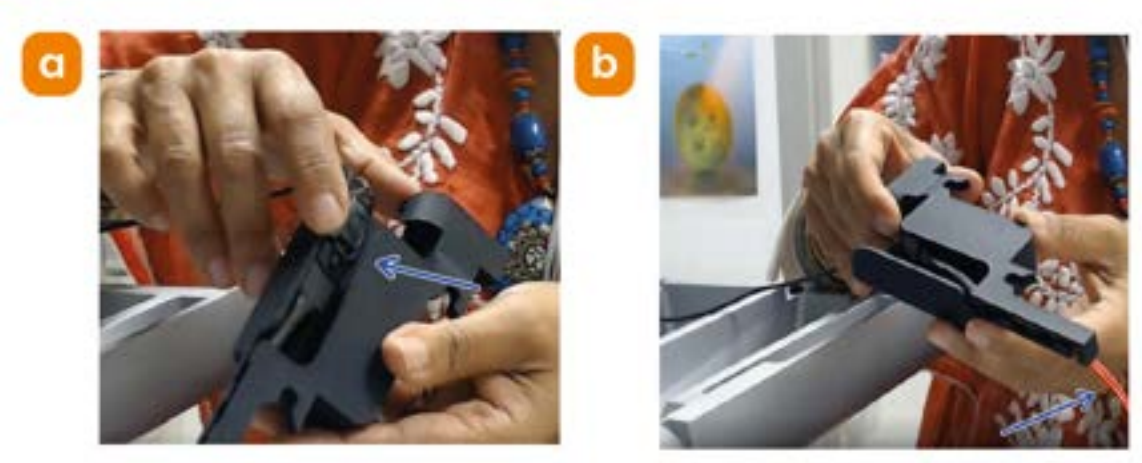


Figure 60: a) Fit the terminal end of the objective heater cable connector into the objective heater cable holster. b) The orange cable leading to the wrap-around heater should hang out of the shaft end of the holster. Now reinsert the holster back into the front chassis of the BC43.

8. Secure the objective heater band to the immersion objective lens, see Figure below.

Figure 61: Secure objective heater band to immersion objective lens. a) Position the brown heater band within the blue velcro strap such that there remains about one fingernail size of space between the ends of the two straps. b) Feed one of the velcro strap into the black buckle on the other end to make a loop. Loosely pull the band loop over the immersion objective lens to its mid-section, then pull on the green velcro strap to tighten around the objective and press down the free end of the tightened strap. c) The orange cables of the objective heater should comfortably be slackened and feeding to the shaft of the objective heater cable holster.

9. Connect the incubator and objective heater to the controller. Please see the Okolab support page and find the user guide for your incubator model. The user guide will provide a connection diagram to support the final steps of the installation.

10. Place the sample in the incubator chamber, the sample holder is built into the Okolab incubator. Add the incubator on top of the stage.
11. Activate the Incubator sample holder option in Fusion, the check box is underneath the objectives display.

Figure 62: Activate Incubator sample holder option in Fusion.

Warning
Failure to activate the incubator option in Fusion may result in the objective lens colliding with the bottom of the stage-top incubator when alternating between different objectives.

12. Remember to turn off the incubator option when using a standard or multi-well XY sample holder.

4.13.2 Using an Oil Immersion Objective Lens with the Okolab Incubator

1. First select one of the instrument's dry/air objective lenses to sight the sample and locate a region of interest to be examined with the oil immersion objective lens. Once a region of interest is found, in Fusion click on the oil objective lens icon (see "a) Click on the oil objective lens icon. b) A message prompt will appear confirming the selection to switch to an oil immersion objective. Click 'Yes' to continue." below).

Figure 63: a) Click on the oil objective lens icon. b) A message prompt will appear confirming the selection to switch to an oil immersion objective. Click 'Yes' to continue.

2. The dry/air objective will disengage from the sample and the chosen oil immersion objective lens will rotate into the active position. Wait for the 'Adding Oil' message prompt to appear on the screen, then lift open the top cover lid ("Adding Oil message" below).

Figure 64: Adding Oil message

3. Lift out the stage-top live-cell imaging chamber and carefully set it aside with the wires still connected on the upper-left corner of the XY stage as shown in "a) and b) remove Okolab incubator. c) Apply a small drop of immersion oil onto the top lens of the oil immersion objective lens." below.

Figure 65: a) and b) remove Okolab incubator. c) Apply a small drop of immersion oil onto the top lens of the oil immersion objective lens.

4. Carefully re-insert the stage-top live-cell imaging chamber into the XY stage.
5. In the Fusion interface, now click the 'Complete' button in the Adding Oil message prompt. The oil immersion objective lens will re-engage the sample coverslip.

Figure 66: a) Adding Oil message prompt. b) An oil drop icon will appear over the oil immersion objective lens icon in the Fusion interface to signify that the oil objective lens is engaged with the sample.

6. Refine the sample focus and XY stage position using the live preview and then continue imaging.
7. When imaging with the oil immersion objective lens is completed, select and click one of the dry/air objective lens icons in the Fusion interface.
8. A prompt requesting confirmation to switch away from the oil objective lens will appear ("a) Confirmation request to switch away from an oil objective. b)

'Cleaning Oil' prompt message." on the next page). Click the 'Yes' button. The oil immersion objective lens will disengage from the sample. Next, the 'Cleaning Oil' prompt message will appear.

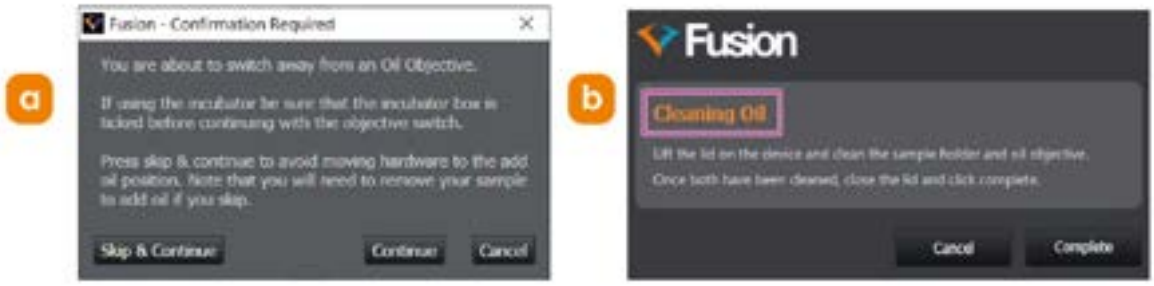


Figure 67: a) Confirmation request to switch away from an oil objective. b) 'Cleaning Oil' prompt message.

9. Lift out the stage-top live-cell imaging chamber and wipe off the oil on the underside of the imaging chamber coverslip.
10. Carefully set the imaging chamber aside with the wires still connected on the upper-left corner of the XY stage as shown in the Figure below.

Figure 68: a) Carefully set the imaging chamber aside. b) Use a solvent-soaked cleaning swab or lens tissue to wipe the oil from the oil immersion objective lens. c) Carefully re-insert the stage-top live-cell imaging chamber into the XY stage.

11. Click the 'Complete' button in the 'Cleaning Oil' prompt message to confirm the removal of oil from the oil immersion objective lens and the sample.

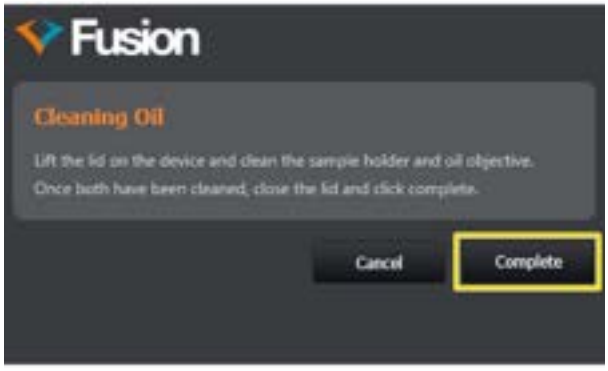


Figure 69: Click the 'Complete' button in the 'Cleaning Oil' prompt message to confirm the removal of oil from the oil immersion objective lens and the sample.

12. The chosen dry/air objective lens will then engage and refocus onto the sample at this position.
13. Refocus the sample and adjust the XY stage position, then continue imaging or shut down Fusion if the imaging session is finished.

4.13.3 Chamber Self Calibration

The goal of the chamber calibration is to automatically adjust the temperature offset of the base and lid of the incubation chamber to maintain the sample at the desired temperature (e.g. 37°C) during imaging.

Figure 70: a) Insert the free green temperature sensor wire into one of the chamber's dedicated openings on the side. b) Secure the green sensor in the desired sample carrier using tape and fill with distilled water. If using a multi-well plate, the green sensor must be placed in a central well and you must fill all the wells and the space in between them.

Figure 71: a) From the controller touch screen, tap Settings > Chamber/Adapter. b) Select the correct insert by tapping the screen until the correct insert appears. Tap OK.

Figure 72: a) Go back to the home-page, and b) set the temperature to 37.0°C

Figure 73: a) Tap Settings > Control Mode > Chamber > Save. b) When the calibration dialog appears, tap on Calibrate.

Figure 74: a) The system will automatically start the calibration procedure and show a progress bar, calibration may take 1-2 hours to complete. b) Once the self-calibration procedure is complete, the window shown will be displayed.

Objective Heater Calibration (optional)

The goal of the heater calibration procedure is to adjust the objective heater temperature while maintaining the sample at the desired temperature (e.g. 37.0 °C), once the immersion media is in contact with the bottom of the well.

Note
This step is optional and should be performed only if using an immersion objective (oil or silicone).

Figure 75: a) Keep the petri dish with the green sample probe in the chamber.Check that the objective heater has been correctly connected to the temperature control unit and secured to the objective of choice. b) Check that the objective heater is enabled by

clicking on Settings > Temp > Objective Heater. If it was not enabled, allow at least 30 mins to equilibrate using the factory settings.

Figure 76: a) Tap on Setting > Temperature > Objective Heater and Calibrate. Then Start. b) The calibration procedure typically takes more than one hour. When the calibration is in process, a reminder icon will appear on the Temperature tab. Once completed, the icon will disappear.

Section 5 - Operation

5.1 Emergency Mains Disconnection

In case of emergency, the disconnecting point of the equipment is the mains power cord connected to the external power supply, or the mains socket switch.

Warning
Switch off the power at the mains socket and remove the mains lead from the external power supply.

5.2 Power up Sequence

1. Turn on BC43 using the switch at the back of the unit.

Figure 77: a) and b) Turn the power switch located on the lower-right corner of the back panel to the ON position. BC43 will undergo its start-up sequence. c) The LED on the front panel will flash a blue light. Once the blue LED light is stable BC43 is ready to be connected to the Fusion control software.

5.3 Power Down Sequence

1. Save any open and altered images.
2. Close the Fusion software interface. Closing the software will put the BC43 instrument into its standby mode.

5.4 Risk Mitigation

Please see "Safety and Warning Information" on page 2 for more information on risk mitigation.

5.5 Standard Procedures

In the following sections additional procedures are described for some routine operations.

5.6 Tools List

Below is a list of tools that are required for procedures described in this user guide. If tools are required they will be listed in the table at the beginning of the instructions for the procedure. Tools are available to order from Andor, please contact your local sales representative referencing the order codes provided below.

Tool	Part code
Immersion oil	**SV-INS-SRVC-TOOLS-19**
Cleaning swabs	**SV-INS-SRVC-TOOLS-14**
Lens tissues	**SV-INS-SRVC-TOOLS-18**
T20 T-shape key	**SV-INS-SRVC-TOOLS-20**
T10 T-shape key	**SV-INS-SRVC-TOOLS-55**
Nitrile gloves	**SV-INS-SRVC-TOOLS-25**
Rocket air blower	**SV-INS-SRVC-TOOLS-17**
Flat head screwdriver	**SV-INS-SRVC-TOOLS-24**
Z-scan sample slide	**XS-ZSCAN-SMPL**
Microfiber cloth	**SV-INS-SRVC-TOOLS-26**
Bubble spirit level	**SV-INS-SRVC-TOOLS-22**
1.5 mm hex driver	**SV-INS-SRVC-TOOLS-41**
Sparkle lens cleaner	**SV-INS-SRVC-TOOLS-15**
Convallaria Montage Sample Slide	**XS-MONT-SMPL**
BC43 Install Qualification slide pack	**XS-SMPL-TEST-SET**

5.7 Inserting/Removing the Sample Holder

Read before starting procedure	
When should the procedure be performed?	The sample holder will need to be removed to install microscope objectives. The sample holder is not seated properly. Putting in a different sample holder
Operation difficulty	Very easy
Time to complete	<1 min
Tools	None
Laser safety considerations	**Class 2 laser radiation is accessible with the lid open at the microscope objective lens or objective lens port, but only one laser can be operational, see "Operational laser access. a) Sample holder and b) Focus Seek and Lock opening. " on page 17.**
Mechanical safety considerations	None

1. The unit can be powered on for this procedure.
2. Remove the sample holder.

Figure 78: a) Grab hold of the front edge of the sample holder with two hands. b) Pull up and away to release the sample holder from the stage. c) Fully remove and set aside the sample holder.

3. Replace sample holder.

Figure 79: a) Push in the sample holder down onto the back edge of the stage opening above the objective lens turret at a roughly 30 degree angle. b) and c) While pressing against the back edge of the sample holder, now push straight down the front edge and left side of the sample holder until it clicks into place securely.

5.8 Adjusting the Sample Holder for Sample Tilt

Read before starting procedure	
When should the procedure be performed?	The first part of these instructions will cover how to determine if the sample holder tilt is good. The following procedure should be performed when the level of tilt in the sample is poor. Poor sample tilt can easily be viewed in widefield or confocal imaging mode with thin specimens and manifests as an uneven change or sweeping gradient in intensity as the microscope focus is scanned through the specimen. System tilt can be caused by one or more of three possible sources: • The levelling screws are not in a neutral state and inadvertently inducing a tilt in the sample. • Some source of tilt has been induced onto the sample; For example, labels on the slide that cause one side of the sample to rest higher in the insert than the other, or the sample is not sitting flat in the insert, etc. • The sample itself, although mounted properly onto the slide with a coverslip, is inherently tilted and the levelling screws must be adjusted to counterbalance the inherent tilt to remove the uneven focus.
Operation difficulty	Moderate
Time to complete	20-30 mins
Tools	1.5 mm hex key 1.5 mm hex driver Multi-channel Gatta-Cells 4C sample slide See "Tools List" on page 100.
Laser safety considerations	**Class 2 laser radiation is accessible with the lid open at the microscope objective lens or objective lens port, but only one laser can be operational, see "Operational laser access. a) Sample holder and b) Focus Seek and Lock opening. " on page 17.**
Mechanical safety considerations	None

Check Sample Tilt

1. The unit can be powered on for this procedure.
2. Insert a monolayer sample e.g. Multi-channel Gatta-Cells 4C sample or a specimen of similar monolayer thickness into the sample holder.
3. In the Fusion Preferences Navigation tab, set the smallest z-step size to 0.1 µm.
4. In Fusion, select the 40x objective lens begin a live preview and focus on the specimen using the EPI channel to view microtubules or actin in the cells. Adjust laser power and exposure time as required to achieve an image with good signal-to-noise ratio. Note that the procedure described here uses the 40x dry objective lens, but for measurements using higher-power objective lenses, the sample tilt should be inspected beforehand and the procedure should be carried out using the chosen objective.
5. Now switch to the corresponding Confocal channel to view microtubules or actin.
6. Focus the sample and switch to the 488 confocal channel.
7. Move through the Z plane using the joystick in 'super fine' mode. Does the whole field of view come into focus at the same position in the Z plane? If the sample all comes into focus in the same Z plane as shown in the screenshot "Good sample tilt." on the next page, then the sample holder is level.

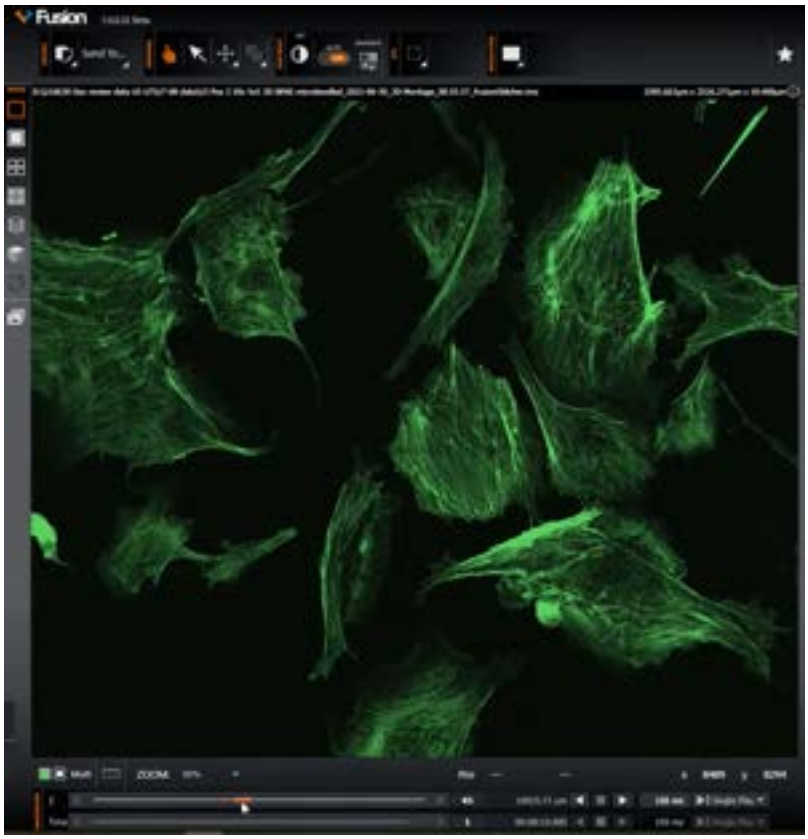


Figure 80: Good sample tilt.

8. If different areas of the sample come into focus at different positions in the Z plane as shown in "Poor sample tilt. a) Z plane: 20. b) Z plane: 25. Different areas of the monolayer sample come into focus at different positions in the Z plane." on the next page, then the stage sample holder will need to be levelled.

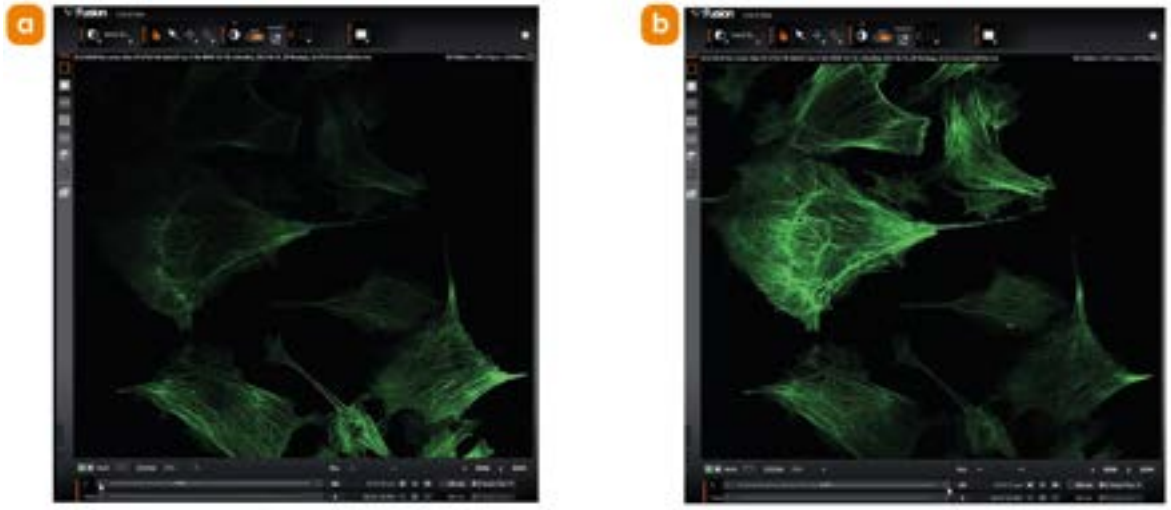


Figure 81: Poor sample tilt. a) Z plane: 20. b) Z plane: 25. Different areas of the monolayer sample come into focus at different positions in the Z plane.

Note
Good and poor sample tilt can also be identified by continuous or discontinuous changes in intensity, respectively, at the borders of tiles in a quick confocal mode montage viewed using Fusion's multiple image display option. See "a) Montage button in Fusion in remote panel. b) Poor sample tilt. c) Good sample tilt." below

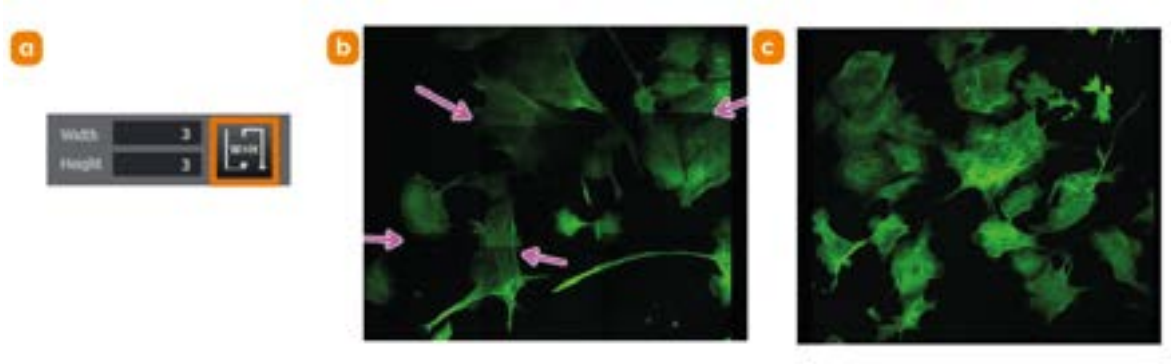


Figure 82: a) Montage button in Fusion in remote panel. b) Poor sample tilt. c) Good sample tilt.

9. Remove the sample holder using the procedure described in "Inserting/Removing the Sample Holder" on page 101.

10. If from the steps above the sample tilt is poor **first** restore the sample holder to a neutral position.

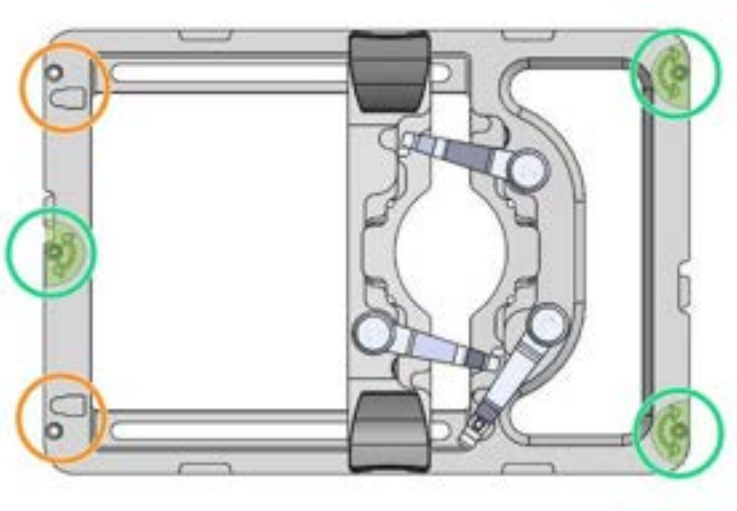


Figure 83: Using the 1.5 mm hex key or 1.5 hex key driver, retract all the levelling setscrews of the sample holder- both the levelling setscrews (green) and the lock setscrews (orange).

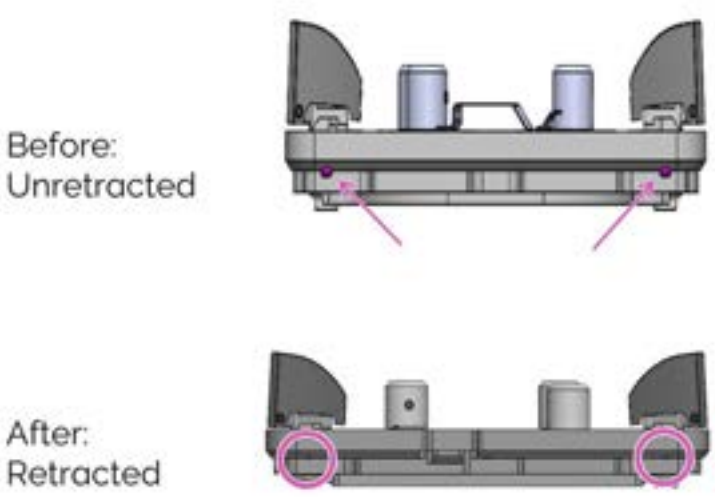


Figure 84: Unscrew (anti-clockwise) such that the bottom tips of the setscrews do not extend below the underside of the base.

11. Repeat steps 3-8 above to check if the source of sample tilt has been resolved, if source of tilt has not been resolved, proceed to the next step.

There are three possible sources of sample tilt, please check in the following order:

1. "Slide labels" below
2. "Sample slide landing area" on the next page
3. "Adjusting the Sample Holder for Sample Tilt" on page 110

Then repeat steps 3-8 above to check if the source of sample tilt has been resolved, before moving to the next possible correction for sample tilt.

Slide labels

1. Check if any labels or stickers have been added to the slide on the same side as the coverslip (i.e. the side which faces down when mounted). Any such label can create a tilt effect. This tilt can become especially visible in larger image montages. Removing these labels and replacing them on the upper side of the slide will improve the levelling of the system.

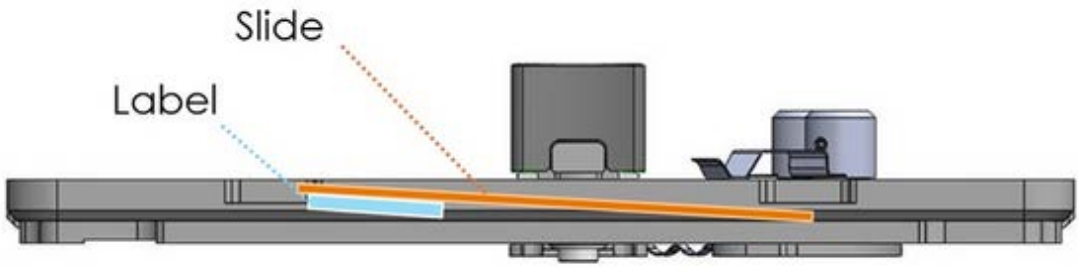


Figure 85: Label causing sample slide tilt.

2. It is also worthwhile to confirm that the sample is correctly mounted on the landing surfaces of the sample holder and to check that the insert is correctly mounted on the landing surfaces of the XY-stage rectangular aperture and secured with the aluminum clips. See "Add a Sample" on page 71.
3. Check if the sample tilt has been corrected using steps 3-8 in "Check Sample Tilt" on page 104
4. If the sample tilt is still poor please proceed to "Sample slide landing area" on the next page

Sample slide landing area

1. The figure highlights the landing surface of the sample holder intended for the sample slide edges.

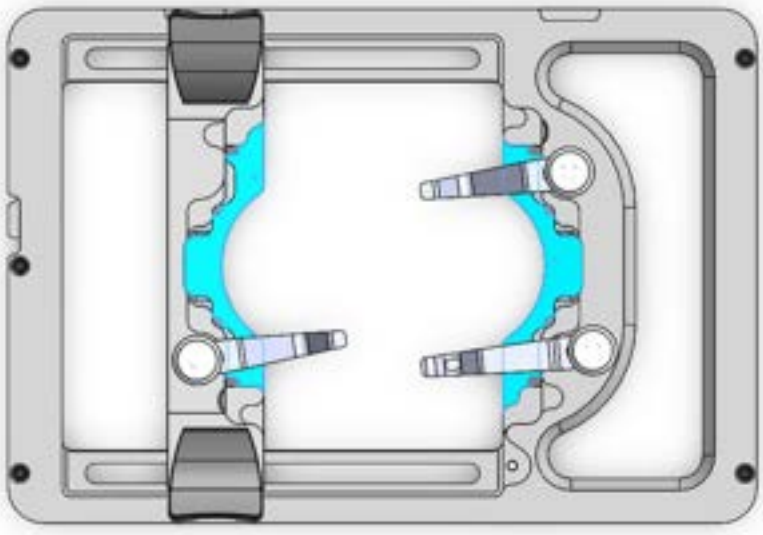


Figure 86: Sample landing area highlighted in blue.

2. To ensure a flat landing surface for the sample slide, check that this area is clean and free of any debris which may create an uneven surface. The area can be cleaned using a cleaning swab when required.
3. Check if the sample tilt has been corrected using steps 3-8 in "Check Sample Tilt" on page 104
4. If the sample tilt is still poor please proceed to "Adjusting the Sample Holder for Sample Tilt" on the next page

Adjusting the Sample Holder for Sample Tilt

1. Retract XY stage insert levelling lock setscrews.

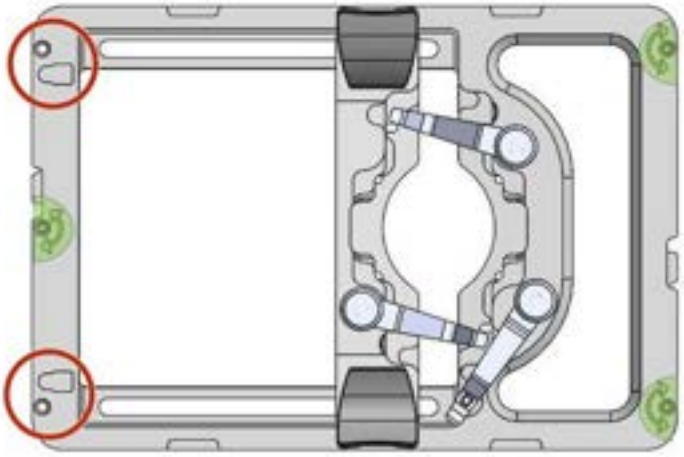


Figure 87: Using the 1.5mm Hex Key or 1.5 mm Hex Driver, first retract the levelling lock setscrews of the XY stage insert by unscrewing (anti-clockwise - ACC) the two indicated screws such that the bottom tips of the setscrews do not extend below the underside of the insert base.

2. Using the 1.5 mm Hex key, ensure all three screws are screwed down such that they stick out ~1 mm below the underside of the sample holder, see "a) Stage sample holder screw positions for adjusting tilt. b) Screw positions on the stage sample holder highlighted. c) Close up of screw protrusion of 1 mm below sample holder." on the next page This allows adjustment up and down at all three points.

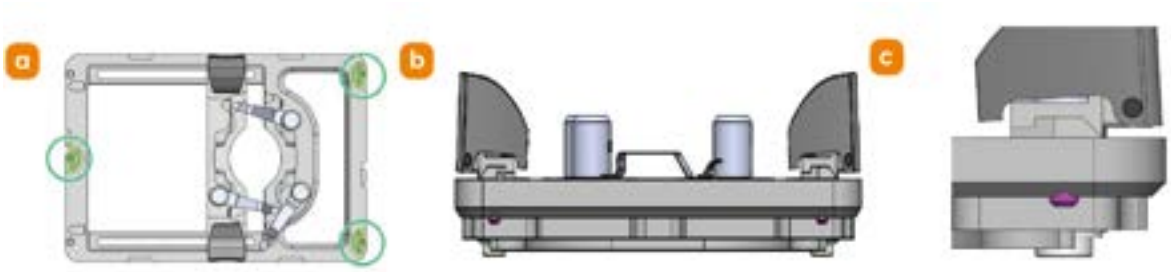


Figure 88: a) Stage sample holder screw positions for adjusting tilt. b) Screw positions on the stage sample holder highlighted. c) Close up of screw protrusion of 1 mm below sample holder.

3. Reinsert the sample holder using the procedure described in "Inserting/Removing the Sample Holder" on page 101.

4. Y adjustment: Check y-axis tilt direction. If you see a brightness sweep in the top-bottom orientation "a) Screw positions on the stage sample holder that impact y-axis tilt direction. b) Image within Fusion interface indicating possible axes of tilt." below) this indicates that levelling is required in the y-axis.

5. Adjust y-axis screws according to tilt along this direction. Start by turning the adjustment screw by half a turn and monitor how the tilt is affected, see "a) Screw positions on the stage sample holder that impact y-axis tilt direction. b) Image within Fusion interface indicating possible axes of tilt." below for screw positions. The front right screw (green) allows the front edge of the sample holder to be raised/lowered. If the sweep is from bottom to top, then turn the green screw clockwise by 1/2 a turn. The back right screw (orange) allows the back edge of the sample holder to be raised/lowered. If from top to bottom, then turn the orange screw clockwise by 1/2 a turn.

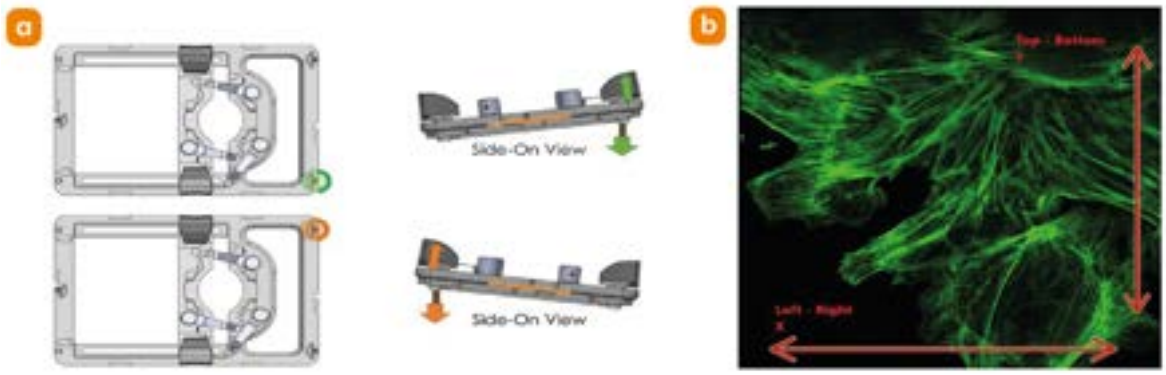


Figure 89: a) Screw positions on the stage sample holder that impact y-axis tilt direction. b) Image within Fusion interface indicating possible axes of tilt.

6. Once an adjustment is made press down firmly on the sample holder at the three screw points. Note the adjustment may cause the sample to move out of the focus plane, if so readjust the focus to find the sample again.

7. If there is still a sweep in the same direction as before then continue adjusting the same screw in clockwise half-turns until it is level or the direction of the

sweep reverses.

- If the sweep is now running top to bottom, then turn the green screw anticlockwise by 1/4 turn.
- If the sweep is now running bottom to top, then turn the orange screw by 1/4 turn.

8. **X adjustment**: Check x-axis tilt direction. If you see a brightness sweep that sweeps in the left to right orientation ("a) Screw positions on the stage sample holder that impact x-axis tilt direction. b) Image within Fusion interface indicating possible axes of tilt." below) this indicates that levelling is required in X.
9. **Only adjust the central green screw indicated in "a) Screw positions on the stage sample holder that impact x-axis tilt direction. b) Image within Fusion interface indicating possible axes of tilt." below**. The central screw (green) allows the left edge of the sample holder to be raised/lowered. Start by turning the central screw (green) by half a turn and monitor how the tilt is affected. If you run out of adjustment with the green screw, then adjust the orange screws, simultaneously and by the same amount. By turning both right-hand side screws (orange) by the same amount the right edge of the sample holder can be raised/lowered.
10. Once an adjustment is made press down firmly on the sample holder at the three screw points, adjust the focus upwards to find the sample again.

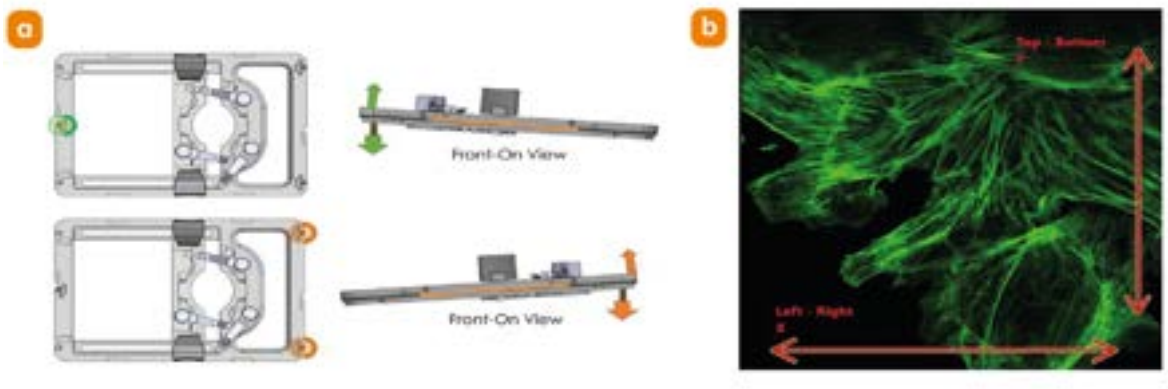


Figure 90: a) Screw positions on the stage sample holder that impact x-axis tilt direction. b) Image within Fusion interface indicating possible axes of tilt.

11. If there is still a sweep in the same direction as before then adjust the same screw in clockwise halfturns until it is level or the direction of the sweep reverses.
12. Recheck x-axis tilt and repeat as required until flat. Lastly recheck the y-axis tilt to ensure this has not been impacted during x-adjustment.
13. Once the stage sample holder is levelled the whole sample should come into focus in the same Z plane as demonstrated in "Good sample tilt." on page 105.
14. When you are satisfied the sample is level you must lock the holder, see "While you observe an in focus Confocal image of a thin sample (e.g. BPAE) slowly screw down (clockwise rotation) the two lock screws." below As you notice the image begin to be affected by the screw touching the metalwork - stop turning the locking screw. When you have done this for both lock screws the sample holder is now locked. The levelling locking setscrews provide extra stability for the stage insert while it rests on the XY stage opening ridge. The insert now has five points of contact against the ridge rather than three, and pressing down on the corners of the stage insert will not alter of the sample stage insert tilt.

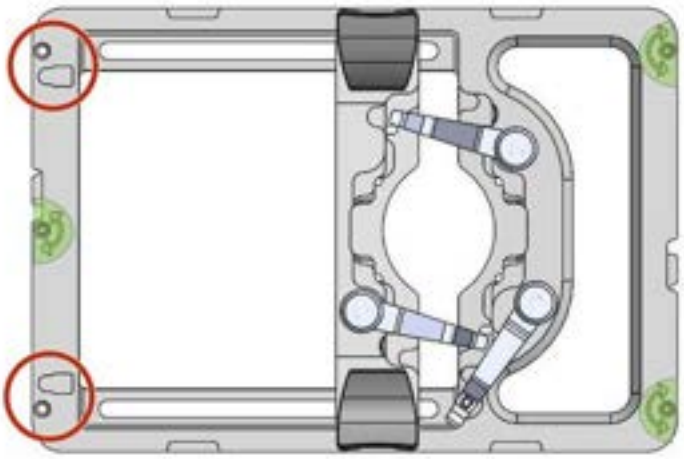


Figure 91: While you observe an in focus Confocal image of a thin sample (e.g. BPAE) slowly screw down (clockwise rotation) the two lock screws.

5.9 Adjusting the Multiwell Plate Holder

Read before starting procedure	
When should the procedure be performed?	The multiwell plate holder will need to be adjusted if a non-standard multiwell plate is used. Please see "Technical Specifications" on page 215 for a full list of compatible multiwell plates that do not require adjustment.
Operation difficulty	Very easy
Time to complete	5 mins
Tools	1.5 mm hex driver See "Tools List" on page 100.
Laser safety considerations	None
Mechanical safety considerations	None

1. The multiwell plate holder has three ball plunger screws to control the size of plate that is acceptable to the slot ("Locations of screw positions on multiwell plate holder for adjustment." below). Using a 1.5 mm hex driver, back the screws off until the base of the ball sits flush with the interior wall of the frame. Note that these setscrews use a threaded lock mechanism that resist movement. This thread resistance will subside slightly once the setscrews start to rotate.

Figure 92: Locations of screw positions on multiwell plate holder for adjustment.

2. With the multiwell plate holder on a flat surface, position the wellplate into the frame with well A1 positioned in the top-left when looking down. Gently

push and hold the side of the multiwell plate to bias it towards the bottom-right corner of the insert slot. While still pushing against the sides of the multiwell plate, turn in the three adjustable setscrews with the 1.5 mm hex driver, until the ball end of each setscrew is in light contact with the side wall of the inserted multiwell plate.

Figure 93: a) Multiwell plate inserted in multiwell plate holder with A1 well in top-left position. b) and c) While still pushing against the sides of the multiwell plate, turn in the three adjustable setscrews with the 1.5 mm hex driver until the ball end of each setscrew is in light contact with the side wall of the inserted multiwell plate.

3. With the wellplate located in the multiwell frame gently grab the wellplate and lift up. If this action also lifts the multiwell frame the assembly is complete. If the multiwell plate separates from the multiwell frame repeat the previous step to adjust.

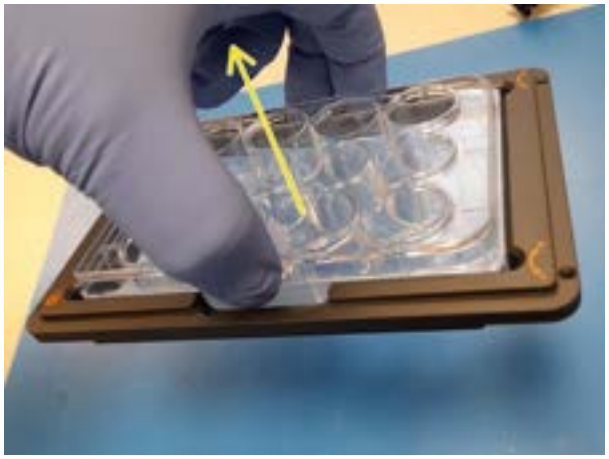


Figure 94: With the wellplate located in the multiwell frame gently grab the wellplate and lift up. If this action also lifts the multiwell frame the assembly is complete.

4. Like the sample holder, the multiwell plate holder has adjustable insert levelling setscrews. Level the multiwell plate stage insert by following the instructions of: "Adjusting the Sample Holder for Sample Tilt" on page 103. The levelling of the insert should be checked and adjusted if necessary,

whenever critical stage-scanning (montage or multi-field) protocols are acquired.

Figure 95: Positions of adjustment screws for levelling the multiwell plate holder.

5.10 Install an Objective Lens

Read before starting procedure	
When should the procedure be performed?	The following procedure describes how to install new objective lenses to the microscope. If you are adding/replacing an objective that was purchased as part of your order please see "Add Objective Lenses to the Microscope" on page 68
Operation difficulty	Very easy
Time to complete	5 mins
Tools	None
Laser safety considerations	**Class 2 laser radiation is accessible with the lid open at the microscope objective lens or objective lens port, but only one laser can be operational, see "Operational laser access. a) Sample holder and b) Focus Seek and Lock opening. " on page 17.**
Mechanical safety considerations	None

Warning
The following procedure should only be carried out using approved BC43 objective lenses. A complete list of supported objectives for this product is available here: "Accessories" on page 43

1. Power ON the BC43 instrument. See "Power up Sequence" on page 99 for further guidance.
2. Run Fusion software.
3. Locate the objectives list from the remote panel and then activate the objective lens/port to be exchanged/upgraded and wait for the unit to rotate in the objective nosepiece to the selected port.

Figure 96: Objective selection in Fusion.

4. Click on the pencil icon to edit the objective lens description.

Figure 97: Enter the objective lens magnification (Mag), numerical aperture (NA), and type (Dry or Oil) of the new objective lens. An objective lens' magnification and numerical aperture can always be checked and verified in the engravings on the side of the objective lens barrel that is being installed.

5. Physically remove/insert the upgraded/exchanged objective lenses as per the instructions of "Add Objective Lenses to the Microscope" on page 68
6. If an objective lens is being removed from the system without exchange, select the pencil and under Type select Unknown. Cap the empty nosepiece position with a carousel cap.

Note
Objectives provided as part of your order will be calibrated for Parfocality and Parcentricity. However, to install replacement or additional objectives, an additional procedure detailed in the Fusion software user guide is required for Parfocality and Parcentration Calibration. The maximum number of objective lenses that can be installed on a BC43 system is 5.

5.11 Add/Remove Immersion Oil to the Oil Immersion Objective Lens

Read before starting procedure	
When should the procedure be performed?	When using the oil objective lens this procedure will need to be performed to add oil.
Operation difficulty	Very easy
Time to complete	5 mins
Tools	Immersion oil, Sparkle lens cleaner, cleaning swabs and lens tissues. See "Tools List" on page 100.
Laser safety considerations	**Class 2 laser radiation is accessible with the lid open at the microscope objective lens or objective lens port, but only one laser can be operational, see "Operational laser access. a) Sample holder and b) Focus Seek and Lock opening. " on page 17.**
Mechanical safety considerations	None

1. Power ON the BC43 instrument. See "Power up Sequence" on page 99 for further guidance.
2. Run Fusion software.
3. Lift open top cover.
4. Locate the sample region of interest using a dry/air objective lens.

Figure 98: a) Select one of the instrument's dry/air objective lenses to sight the sample and locate a region of interest to be examined with the oil immersion objective lens. b) Once a region of interest is found, in Fusion click on the oil objective lens icon. c) A message prompt will appear confirming the selection to switch to an oil immersion objective. Click 'Yes' to continue.

5. Wait for the ‘Adding Oil’ message prompt to appear on the screen, then lift open the top cover lid.

Figure 99: Adding oil message in Fusion interface.

6. Apply immersion oil to oil immersion objective lens

Figure 100: a) The oil immersion object lens will be accessible to apply oil near the upper-right corner of the stage insert. b) Add a drop of immersion oil to the objective. c) Now click the ‘Complete’ button in the Adding Oil message prompt.

7. Once the hardware has finished moving, see next figure, refine the sample focus and XY stage position using the live preview and then continue imaging.

Figure 101: a) The 'Moving Hardware' message prompt will appear, and the oil immersion objective lens will disengage and the XY stage will move the specimen back to its previous position viewed by the dry/air objective lens. b) Next, the oil immersion objective lens will raise to make the oil drop contact the sample coverslip. c) An oil drop icon will appear over the oil immersion objective lens icon in the Fusion interface to signify that the oil objective lens is engaged with the sample.

8. Once imaging is completed, clean and remove oil from oil immersion objective lens. First select and click one of the dry/air objective lens icons in the Fusion interface.

Figure 102: a) A prompt message appears requesting confirmation to switch away from the oil objective lens. Click the 'Yes' button. b) The 'Cleaning Oil' prompt message will appear; the oil immersion objective lens will disengage from the specimen and the XY stage will move back to the oil application position. c) Use a solvent-soaked cleaning swab or lens tissue to wipe the oil from the oil immersion objective lens.

9. Confirm oil removal and continue imaging with dry objective lens.

Figure 103: a) Click the 'Complete' button in the 'Cleaning Oil' prompt message to confirm the removal of oil from the oil immersion objective lens and the sample. b) The 'Moving Hardware' prompt message will appear and the XY stage will return to the previous sample position. The chosen dry/air objective lens will then engage and refocus onto the sample at this position. c) Refocus the sample and adjust the XY stage position, then continue imaging or shut down Fusion if the imaging session is finished.

5.12 Objective Correction Collars

Read before starting procedure	
When should the procedure be performed?	Some high magnification objective lenses used with the BC43 possess a correction collar. In most instances the objective correction collar should not need to be adjusted and a setting of 0.17 millimeters will provide optimal image quality. However, there may be certain experiments where enhancing image contrast and minimizing image degradations due to coverslip thickness variations or sample-mounting medium refractive index differences mean that the correction collar may need to be adjusted. For experiments requiring the best possible imaging quality, it is recommended that the objective correction collar, if present, be adjusted before acquiring any images of the specimen. What follows below is a general procedure to attain the optimum correction collar setting for most experimental situations.
Operation difficulty	Moderate
Time to complete	5 mins
Tools	None
Laser safety considerations	**Class 2 laser radiation is accessible with the lid open at the microscope objective lens or objective lens port, but only one laser can be operational, see** **"Operational laser access. a) Sample holder and b) Focus Seek and Lock opening. " on page 17****.**
Mechanical safety considerations	None

1. Mount the specimen with #1.5 glass coverslip that has a standard nominal thickness of 0.17 millimeters and a refractive index of 1.515.

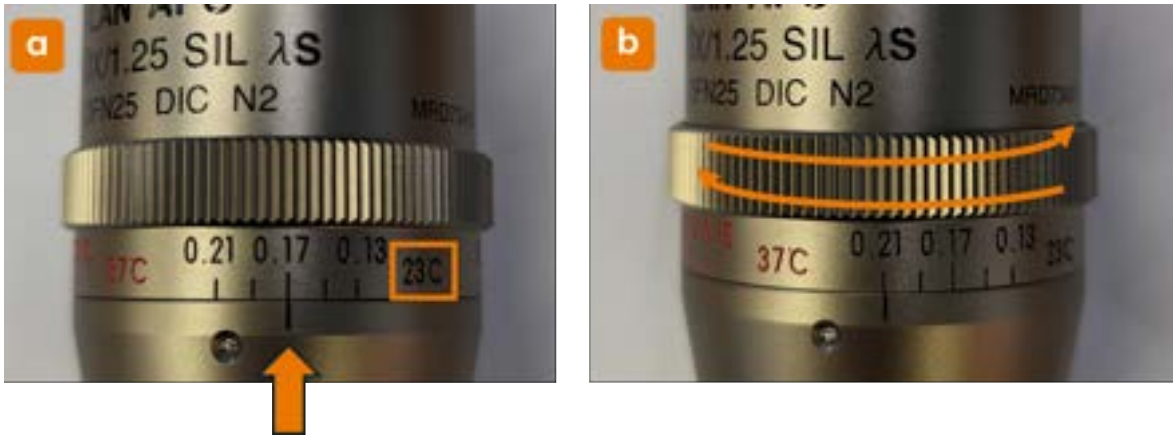


Figure 104: a) Position the correction collar so that the indicator mark on the objective barrel coincides with the 0.17 mm scale mark engraved on the collar housing. b) The collar can be adjusted by rotating in either direction.

Figure 105: If you are imaging your specimen in a 37 degree heated environment such as the Okolabs stage-top incubator (H301-K-FRAME-AD), check if your objective lens has a correction collar scale for this temperature and use this scale instead of the room temperature scale.

2. Mount the specimen onto the microscope stage (see "Add a Sample" on page 71). Bring the sample into focus with the channel of interest onto the camera using the software live-preview. Adjust the laser power and exposure time for good a signal-to-noise ratio as required.

3. Try to locate and center the live preview on a small, bright point-like source of light within the sample or image background. This can be small bit of auto-

fluorescent background or an isolated fluorescent structure within the sample.

4. Slowly move the focus up and down around this point of light a few steps of the software virtual joystick and notice if the out-of-focus image of this point of light looks the same as when focusing upwards compared to focusing downwards.
5. If the out-focus light response does not appear symmetric when focusing up and down, then the correction collar of the objective lens must be adjusted. Remove the specimen to access the correction collar and try rotating the collar indicator a larger compensation value (0.18-0.23) on the scale. Now repeat steps 2-4.
6. If the symmetry of the out-of-focus light around the point source has degraded further, iteratively repeat this procedure by rotating the correction collar in the opposite direction (toward lower or higher compensation values) to find the position maximum focal symmetry around the point source.

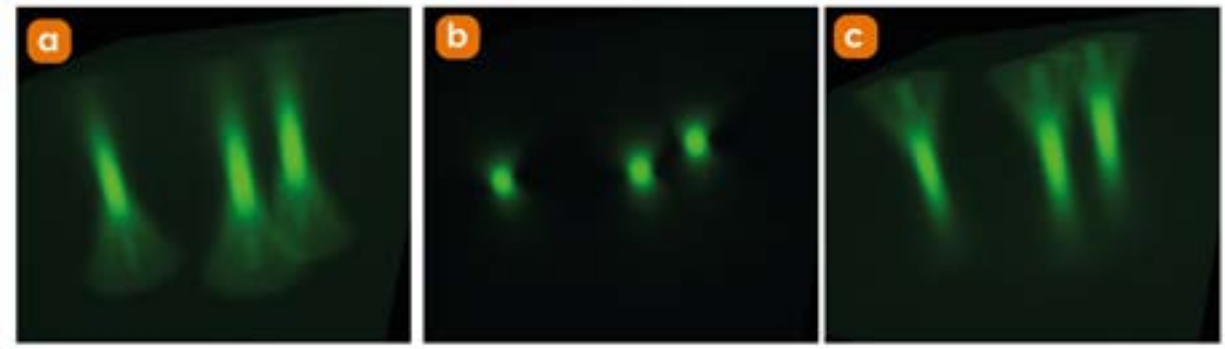


Figure 106: When the correction collar is correctly set to a value that matches the thickness of the sample's coverslip spherical aberrations are reduced and a symmetrical response is observed above and below the sample volume. In the above example a sample containing 1 µm beads mounted on a standard 170 µm coverslip is imaged at 40x to obtain a z-stack with the correction collar set at a) 110 µm b) 170 µm and c) 230 µm. When the correction collar value is set too low (a) or too large (c) the asymmetric nature of the 3D image is clearly visible; when set at the correct value for the sample (b) the best quality image is produced.

5.13 Deconvolution

For information on the principles of deconvolution within Fusion please see the Deconvolution section of the Fusion user guide.

To understand how to perform deconvolution on the BC43 please see performing deconvolution the in Fusion user guide.

Section 6 - Maintenance and User Service Procedures

6.1 Cleaning and Decontamination Procedures

The following sections describe user procedures for maintaining the BC43 instrument including cleaning and decontamination operations.

Tools may be required to perform certain procedures, if required, tools are listed in the table at the beginning of the instructions. Please see order codes for tools available from Andor in the table in "Tools List" on page 100.

6.2 Cleaning Outer Surfaces of BC43 Unit

Read before starting procedure	
When should the procedure be performed?	Outer surface of the BC43 is dirty or dusty.
Operation difficulty	Very easy
Time to complete	5 mins
Tools	Lint free cloth
Laser safety considerations	None
Mechanical safety considerations	None

Ensure the lid of the BC43 is closed. Using a damp lint free cloth e.g. micro fibre cleaning cloth, soaked in warm soapy water, wipe down the outer surfaces of the unit to remove any dust or dirt.

6.3 Cleaning the Drip Tray

Read before starting procedure	
When should the procedure be performed?	If oil or any other liquid appears on the drip tray it can be cleaned.
Operation difficulty	Very easy
Time to complete	5 mins
Tools	Absorbent wipes Nitrile gloves See "Tools List" on page 100
Laser safety considerations	**Class 2 laser radiation is accessible with the lid open at the microscope objective lens or objective lens port, but only one laser can be operational,** **"Operational laser access. a) Sample holder and b) Focus Seek and Lock opening. " on page 17**
Mechanical safety considerations	None

1. Power off the BC43 before starting this procedure.
2. Push to release the front access panel of the BC43.
3. Using any absorbent tissue (for example, KIMTECH Kimwipes) starting from the back left wipe down and around the slope of the tray.

Figure 107: The image shows the motorised objective turret and Z-stage installed in the unit. The drip tray is highlighted in blue.

4. Dispose of used tissue and ensure hands are clean and free from oil before continuing to operate the instrument.

6.4 Cleaning the Objective Lenses

Read before starting procedure	
When should the procedure be performed?	It is good practice to clean microscope objectives regularly.
Operation difficulty	Easy
Time to complete	5 mins
Tools	Lens tissue Sparkle lens cleaner Nitrile gloves See "Tools List" on page 100.
Laser safety considerations	**Class 2 laser radiation is accessible with the lid open at the microscope objective lens or objective lens port, but only one laser can be operational, see "Operational laser access. a) Sample holder and b) Focus Seek and Lock opening. " on page 17.**
Mechanical safety considerations	None

1. Remove the objective lens to be cleaned as per the instructions of "Install an Objective Lens" on page 117.
2. Place a drop of Sparkle Lens Cleaner or equivalent lens cleaning solvent on a single piece of lens tissue.
3. Lay the wet portion of the lens tissue on the top lens of the objective, then slowly pull tissue to the left until the drop streak completely dries.

Figure 108: a) Place a drop of Sparkle Lens Cleaner or equivalent lens cleaning solvent on a single piece of lens tissue. b) and c) Lay the wet portion of the lens tissue on the top lens of the objective, then slowly pull the tissue to the left until the drop streak completely dries.

4. Replace the objective lens back into its correct nosepiece position as per the instructions of "Install an Objective Lens" on page 117.
5. Any oil immersion objective lenses should be cleaned as per the instructions of "Add/Remove Immersion Oil to the Oil Immersion Objective Lens" on page 119.

6.5 Cleaning the Infinity Space Window

Read before starting procedure	
When should the procedure be performed?	If you see artefacts on the image, the front chassis infinity space sealing window may be contaminated and require cleaning.
Operation difficulty	Easy
Time to complete	10 - 15 mins
Tools	Optical swab Nitrile gloves See "Tools List" on page 100.
Laser safety considerations	**The unit must be powered OFF during this procedure.** **If power is re-established while cleaning the infinity space window, Class 2 laser radiation is accessible with the front access panel door open (but only one laser can be operational), see "Operational laser access. a) Sample holder and b) Focus Seek and Lock opening. " on page 17.** **If the instrument is powered OFF, then there is no hazard.**
Mechanical safety considerations	**The unit must be powered OFF during this procedure.** **The FSL module's electrical connection is achieved via a blind mate connection. If power is re-established while replacing the FSL module and a finger is placed over/beyond the module edge and under the drip tray, the Z-stage could potentially home and create a finger crush hazard. Please see "Risk No 3: Focus Seek and Lock Module Replacement" on page 27** **If the instrument is powered OFF, then there is no hazard.**

1. Remove Focus Seek and Lock module, follow instructions in "Replacing the Focus Seek and Lock Module" on page 146.
2. Put a pair of clean rubber gloves.
3. Add a few drops of lens cleaning solvent to an optical swab.
4. Insert the swab in the Focus Seek and Lock module space and position the solvent-soaked swab over the infinity space window.
5. Use a torch to check to window for contamination before and after cleaning.

Figure 109: Starting from the centre of the window, press the swab lightly against the window and spin the swab outwards to the edges of the window in a spiral pattern. Move the swab at an appropriate speed that does not leave behind any solvent streaks on the window.

6. Dispose of the swab when the cleaning is complete. Use a fresh swab, with a few drops of lens cleaning solvent, if it is necessary to clean the window again.
7. Reinsert Focus Seek and Lock module follow instructions in "Replacing the Focus Seek and Lock Module" on page 146.

6.6 Cleaning the Focus Seek and Lock Module

Read before starting procedure	
When should the procedure be performed?	If you see artefacts on the image, the front chassis infinity space sealing window may be contaminated and require cleaning.
Operation difficulty	Easy
Time to complete	10 - 15 mins
Tools	Wera Torx Screwdriver Bit, T20 Wera 1/4 in Hex Adjustable Torque Screwdriver, 1.2-3 Nm Rocket Air Blower Optical Lens Cleaner Cotton swab Nitrile gloves See "Tools List" on page 100.
Laser safety considerations	**The unit must be powered OFF during this procedure.** **If power is re-established while replacing the FSL module, Class 2 laser radiation is accessible with the front access panel door open (but only one laser can be operational), see "Operational laser access. a) Sample holder and b) Focus Seek and Lock opening. " on page 17.** **If the instrument is powered OFF, then there is no hazard.**
Mechanical safety considerations	**The unit must be powered OFF during this procedure.** **The FSL module's electrical connection is achieved via a blind mate connection. If power is re-established while replacing the FSL module and a finger is placed over/beyond the module edge and under the drip tray, the Z-stage could potentially home and create a finger crush hazard. Please see "Risk No 3: Focus Seek and Lock Module Replacement" on page 27.** **If the instrument is powered OFF, then there is no hazard.**

1. Remove Focus Seek and Lock module, follow instructions in "Replacing the Focus Seek and Lock Module" on page 146.
2. Put a pair of clean rubber gloves.
3. Unscrew dichroic mirror mount.

Figure 110: a) Turn the removed Focus Seek and Lock module around to its backside and locate the dichroic mirror mount locking setscrew. b) The orange arrow on the back of the module points to this setscrew. c) Unscrew the mirror mount from the module using the T20 Torx bit loaded into the 1.2-3 Nm Torque Gun.

4. Remove dichroic mirror mount from module. Grab hold of outward facing corner of the dichroic mirror mount as shown and lift it off from the Focus Seek and Lock module.

Figure 111: Grab hold of outward facing corner of the dichroic mirror mount as shown and lift it off from the Focus Seek and Lock module.

5. Clean the front side of Focus Seek and Lock module dichroic mirror. Set the removed dichroic mirror mount on a clean benchtop surface. Blow away any large dust particles from the mirror surface using the Rocket Air Blower.
6. Soak a rigid tip spear swab with lens cleaning solvent (e.g. methanol) and shake off excess solvent. Starting from the mirror's middle, press the swab end gently against the mirror optic and move the swab slowly outwards in a spiral motion to the mirror edges. Dispose of the swab immediately. Tilt up the mirror in the room light and examine for any solvent residue streaks. If any streaks are observed, repeat the above procedure using a freshly solvent-soaked swab.

Figure 112: Set the removed dichroic mirror mount on a clean benchtop surface. Soak a rigid tip spear swab with lens cleaning solvent and shake off excess solvent. Starting from the mirror's middle, press the swab end gently against the mirror optic and move the swab slowly outwards in spiral motion to the mirror edges. Dispose of the swab immediately. Tilt up the mirror in the room light and examine for any solvent residue streaks. If any streaks are observed, repeat the above procedure using a freshly solvent-soaked swab.

7. Flip the dichroic mirror mount over and examine bottom side of the optic (optional). If contamination is observed on this surface, repeat the cleaning procedure described in the previous step with the swab entering through the mount's circular hole.

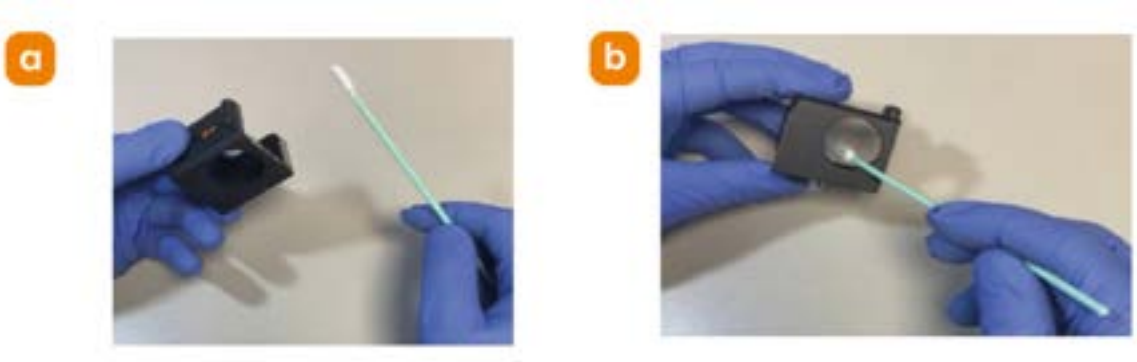


Figure 113: Flip the dichroic mirror mount over and repeat the cleaning procedure described in the previous step with the swab entering through the mount's circular hole.

8. Reattach dichroic mirror mount to module.

Figure 114: Fit the dichroic mirror mount back onto the Focus Seek and Lock module and lock in by tightening the setscrew with the T20 Torx bit loaded into the 1.2-3 Nm Torque Gun.

9. Reinsert Focus Seek and Lock module follow instructions in "Replacing the Focus Seek and Lock Module" on page 146.

6.7 Regular Checks

- The state of the product should be checked regularly, especially the integrity of the PSU and the mains cable.
- Do not use equipment that is damaged.

6.7.1 Minimum Computer Requirements

Please see the specification sheet for most up to date information on computer requirements.

Please note USB 3.0, USB 3.1 (Gen 1) and USB 3.2 Gen 1x1 are equivalent.

6.8 Windows Operating System Updates

Andor recommends that the latest updates to the Windows operating system of the PC workstation be applied when they are available. Windows operating system updates are regularly distributed by Microsoft if the PC workstation is networked to the internet. The system administrator can schedule when these updates are downloaded, installed, and applied. It is possible to delay operating system updates, but not indefinitely. Therefore, it is highly recommended that the user check for and then download and install Windows updates before any critical long-term imaging experiments to prevent Windows from restarting the PC workstation to apply an operating system update that has already been delayed the maximum number of times that is permissible.

6.9 Annual Electrical Safety Checks

- It is advisable to check the integrity of the insulation and protective earth of the product on an annual basis, e.g. U.K. PAT testing. However over time the repetition of dielectric strength tests can damage safety insulation.
- Do not use equipment that is damaged.

6.10 Maintenance Packages

Andor offer a variety of maintenance packages, designed to maximize return on investment, tailored to suit the needs of the customer, ensuring that global expertise is delivered locally.

For more information on the packages available, please visit our website.

6.11 Service Options

A range of services are available for the BC43, these include:

- Installation qualification
- Operational qualification
- Maintenance packages
- Onsite and depot repair
- Spares and consumables

For more information or pricing, please visit our website.

6.12 User Service Procedures

The following sections describe user procedures for self-servicing the BC43 instrument. Please contact our customer support team to assist with troubleshooting and to order replacement parts. Additionally, if you require support to perform any of the operations, please contact our customer support team who will be happy to assist you.

6.13 Open or close the front access panel door

A number of the following procedures will require the front access panel door to be opened before starting the procedure and closed when the procedure is complete.

Push to release on the hinged front panel door to gain access to the inside of the front chassis.

Figure 115: Open front access panel door.

Lift up and close the front access panel and push to "click!" closed.

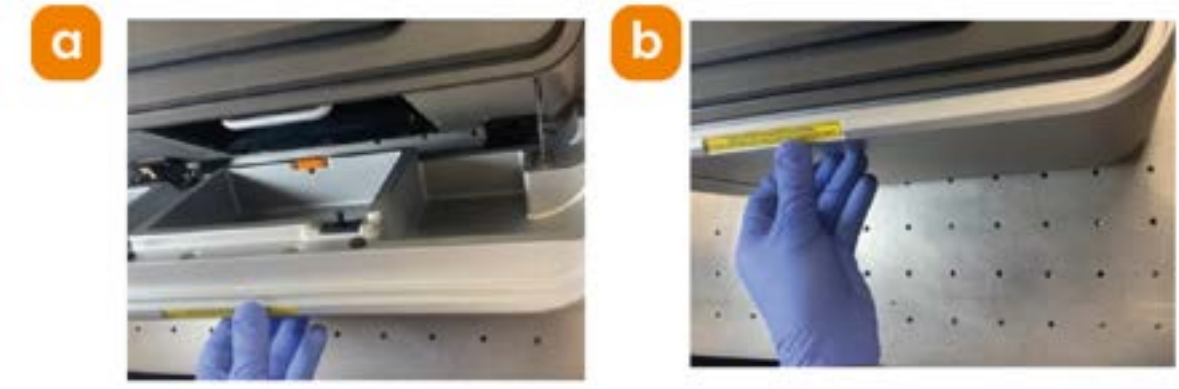


Figure 116: Close the front access panel door.

6.14 Replacing the Focus Seek and Lock Module

Read before starting procedure	
When should the procedure be performed?	If focus stabilisation for protocols or the coverslip auto finder are not working.
Operation difficulty	Easy
Time to complete	15-20 mins
Replacement part codes	Focus Seek and Lock Module **SV-INS-FSL**
Tools	RS PRO size T20 T Shape Long, Short arm Torx Key See "Tools List" on page 100.
Laser safety considerations	**The unit must be powered OFF during this procedure.** **If power is re-established while replacing the FSL module, Class 2 laser radiation is accessible with the front access panel door open (but only one laser can be operational), see "Operational laser access. a) Sample holder and b) Focus Seek and Lock opening. " on page 17.** **If the instrument is powered OFF, then there is no hazard.**
Mechanical safety considerations	**The unit must be powered OFF during this procedure.** **The FSL module's electrical connection is achieved via a blind mate connection. If power is re-established while replacing the FSL module and a finger is placed over/beyond the module edge and under the drip tray, the Z-stage could potentially home and create a finger crush hazard. Please see "Risk No 3: Focus Seek and Lock Module Replacement" on page 27.** **If the instrument is powered OFF, then there is no hazard.**

1. First lift open the BC43 top cover. And open the front access panel door. If you are unsure how to do this please see "Open or close the front access panel door" on the previous page.
2. Unlock the Focus Seek and Lock Module.

Figure 117: Using the T20 Torx bit loaded into the Adjustable Torque Screwdriver 1.2 - 3 Nm, loosen the front captive screw to unlock the Focus Seek and Lock Module.

3. Remove Focus Seek and Lock Module.

Figure 118: a) Grab hold of the Focus Seek and Lock module handle. b) Ensure to use two hands, one at the front and one to support the module as you slowly pull the Focus Seek and Lock Module straight back and outwards until it is completely free of the front chassis.

4. Focus Seek and Lock chassis mating connections.

Figure 119: Note the following features on the underside of the Focus Seek and Lock module: (quantity 2) magnets (quantity 2) guide pins, the module's blind-mate electrical

connector. Observe the corresponding connection landings and socket in the open front chassis.

5. Focus Seek and Lock insertion.

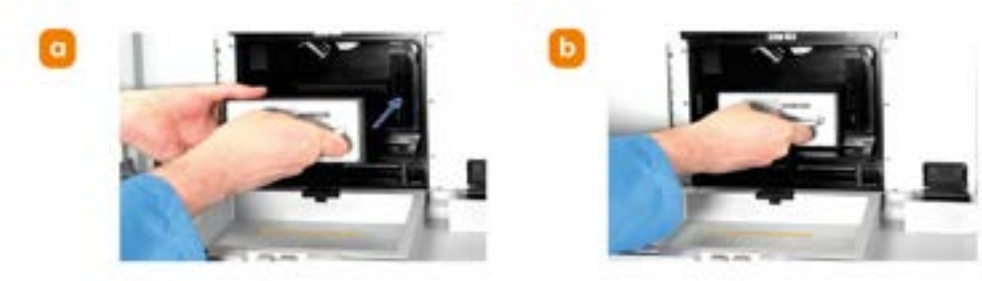


Figure 120: With one hand holding the Focus Seek and Lock module front handle and the other hand holding the front frame of the module, carefully guide the device back into the front chassis. When the replacement module is almost fully inserted, apply slight pressure, and the module pins and magnets should click into place.

6. Focus Seek and Lock lock-in.

Figure 121: Using the T20 Torx driver, tighten the front captive screw to lock the Focus Seek and Lock module into the front chassis. Torque to 2.2 Nm.

7. Lift up and close the front access panel and push to "click!" closed. If you are unsure how to do this please see "Open or close the front access panel door" on page 145. Close the instrument's top lid.

8. FSL calibration. If a new FSL module has been loaded into the instrument, the module must be calibrated in Fusion. It is good practice to check and verify the functionality of the FSL module any time it is removed/replaced from the unit. Failure to calibrate new FSL replacement modules will cause the Find

Coverslip and Focus Stabilization features of the instrument to not function properly.

Note
Customers self-installing a replacement FSL module must make arrangements with their local Andor service representative to remotely calibrate the FSL module in Fusion.

6.15 Replacing the Objective Heater Holster

Read before starting procedure	
When should the procedure be performed?	This procedure is required to install an objective heater jacket, or to replace a broken clip on the holster
Operation difficulty	Easy
Time to complete	10-20 mins
Tools	Objective Heater Holster **SV-INS-CBL-MGT-C**
Laser safety considerations	**Class 2 laser radiation is accessible with the lid open at the microscope objective lens or objective lens port, but only one laser can be operational, see "Operational laser access. a) Sample holder and b) Focus Seek and Lock opening. " on page 17.**
Mechanical safety considerations	None

1. Open the front access panel door. From the top, push to release on the hinged front panel door to gain access to the inside of the front chassis. If you are unsure how to do this please see "Open or close the front access panel door" on page 145.
2. Locate and remove objective heater cable holster.

Figure 122: a) The objective heater cable holster is located on the upper-left corner of the front chassis left wall which is now accessible through the front access panel door opening. b) Grab hold of the lower end of the holster with one hand and pull back towards the front of the instrument to release the holster from its locator pins. c) Then pull the holster to the right to remove from the front access door panel opening.

3. Insert objective heater cable holster. Note the location of the replacement holster clips for the locator pins inside the front chassis. Take hold of the

replacement holster and sneak it into the front chassis against the upper-left corner of the left front chassis wall. Now slide and push forward the holster until it grabs onto and clips around the two locator pins.

Figure 123: a) Note the location of the replacement holster clips for the locator pins inside the front chassis. b) Take hold of the replacement holster and sneak it into the front chassis against the upper-left corner of the left front chassis wall. c) Now slide and push forward the holster until it grabs onto and clips around the two locator pins.

4. Close front access panel door. Lift up and shut the front access panel door and push to "click!" it into place. If you are unsure how to do this please see "Open or close the front access panel door" on page 145.

6.16 Replacing the XY Stage

Read before starting procedure	
When should the procedure be performed?	Please contact the Andor service team prior to starting this procedure if an XY stage fault displays in Fusion.
Operation difficulty	Easy
Time to complete	10 - 20 mins
Replacement part codes	XY Stage **SV-INS-XY-STG**
Tools	Wera Torx Screwdriver Bit, T25 T25 Torx Bit See "Tools List" on page 100.
Laser safety considerations	**The unit must be powered OFF during this procedure.** **If power is re-established while replacing the XY stage, Class 2 laser radiation is accessible with the front access panel door open (but only one laser can be operational), see "Operational laser access. a) Sample holder and b) Focus Seek and Lock opening. " on page 17.** **If the instrument is powered OFF, then there is no hazard.**
Mechanical safety considerations	**The unit must be powered OFF during this procedure.** **The XY stage's electrical connection is achieved via a blind mate connection. If power is re-established while replacing the FSL module and a finger is placed over/beyond the module edge and under the drip tray, the Z-stage could potentially home and create a finger crush hazard. Please see "Risk No 4: XY Stage Replacement" on page 29.** **If the instrument is powered OFF, then there is no hazard.**

1. On the BC43 unit turn the power switch located on the lower-right corner of the back panel to the OFF position.
2. Lift open top cover of the BC43. Grab hold of the orange handle and lift up to open to access the sample stage and objective lens turret.
3. Remove sample holder and objective lenses, cap nosepiece holes.

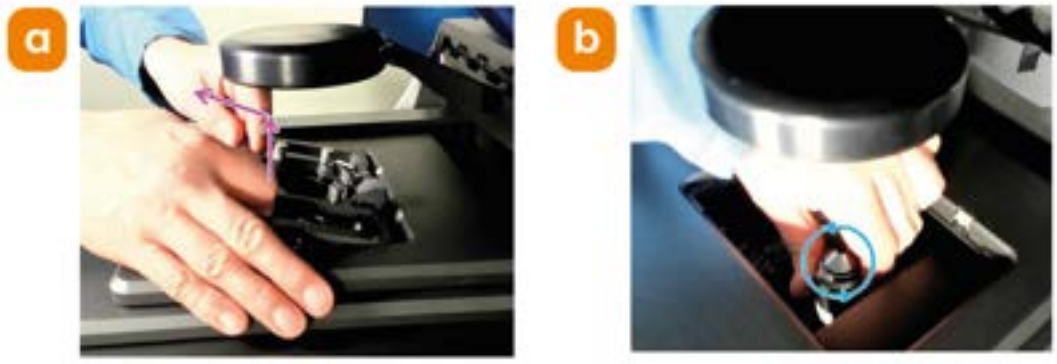


Figure 124: a) Remove sample holder. b) Manually unscrew each objective lens from the nosepiece turret.

4. Pull down front access panel door. From the top, pull down on the hinged front panel door to gain access to the inside of the front chassis. If you are unsure how to do this please see "Open or close the front access panel door" on page 145.
5. Remove XY stage front chassis screws.

Figure 125: a) Location of chassis screws. b) Unscrew first and c) second screws.

6. Attach the T20 Torx bit to the 1.2-3 Nm Torque Gun and completely loosen the rear chassis XY stage captive screw.

Figure 126: a) Location of the captive screw. b) Guide the Torx bit to the captive screw head with a second hand through the stage insert hole on the topside of the stage. c) Loosen rear chassis XY stage captive screw.

7. Lift off and remove stage. Set the removed stage down on a flat, clean surface.

Figure 127: a) With the front screws and rear captive screw backed off, grab hold of the stage with one hand through the sample holder and nudge up the lower left corner to release the stage from the blind mate connectors. When the stage is free of the chassis, carefully pull it away from the unit, being careful not to crash or knock the transmitted light illuminator. b) Note the exposed stage bind mate connectors.

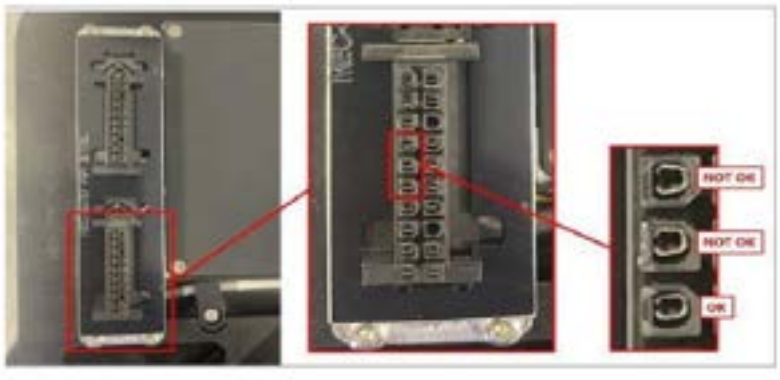


Figure 128: The replacement stage pins must press into these connectors when fitted into the instrument. Check the pins and use a set of tweezers to close the connectors into a "C" shape if necessary (double check that the instrument power is OFF).

8. Note and record the serial number of the new stage to be inserted into the BC43 unit and insert new stage. Take care when reattaching XY stage not to

damage the cable crimps. It should drop into position freely.

Warning
TAKE CARE WHEN REATTACHING XY STAGE NOT TO DAMAGE THE CABLE CRIMPS. IT SHOULD DROP INTO POSITION FREELY. DO NOT APPLY DOWNWARD FORCE ON XY STAGE IF IT HAS NOT SEATED PROPERLY. REMOVE IT AND TRY AGAIN.

Figure 129: a) Grab hold of the new stage to be inserted and guide it under the transmitted light turret and then tilt the back edge downwards into the recess adjacent to the left and right-side cosmetic panels. b) and c) Apply downward pressure onto the back and front corners of the stage, while checking from the side to see if the stage fits into its space and that the stage pins located on the underside of the lower-left corner have made good contact with the blind mate connectors.

Figure 130: Ensure the XY stage is centred by looking at the rear corners where the XY stage meets the rear chassis. Both sides should be flush.

9. Loosely screw in the front XY stage screws using the T25 Torx Bit loaded into AdjTorqGun 1.2 - 3 Nm just enough so that the screws hang in place. **DO NOT TIGHTEN AT THIS STAGE**. Now tighten the rear captive screw, torque set to 3.0 Nm. Now go back and tighten theloose front screws to 3.0 Nm.

Figure 131: a) and b) loosely screw in the front XY stage hex screws (do not tighten at this stage) and c) the rear captive screw using the1.2-3 Nm torque gun. Now tighten the rear captive screw, torque set to 3.0 Nm. Now go back and tighten the loose front screws to 3.0 Nm.

10. Close front access panel door. If you are unsure how to do this please see "Open or close the front access panel door" on page 145.
11. Replace system objective lenses and sample holder. Please see "Add Objective Lenses to the Microscope" on page 68.
12. Close the top cover.

6.17 Replacing Front Access Panel Door

Read before starting procedure	
When should the procedure be performed?	The front access panel door drawbridge mechanism has failed or is broken.
Operation difficulty	Easy
Time to complete	10 - 15 mins
Replacement part codes	Front Access Panel Door **SV-INS-DOOR**
Tools	Nitrile gloves T25 T-Shape Key See "Tools List" on page 100.
Laser safety considerations	**The unit must be powered OFF during this procedure.** **If power is re-established while replacing the front access panel door, Class 2 laser radiation is accessible with the front access panel door and/or the top lid open (but only one laser can be operational), see "Operational laser access. a) Sample holder and b) Focus Seek and Lock opening. " on page 17.** **If the instrument is powered OFF, then there is no hazard.**
Mechanical safety considerations	None

1. Ensure the BC43 unit is powered OFF before starting this procedure. Disconnect the blue USB cable and the power adapter cable from the BC43 unit.
2. Pull down front access panel door. If you are unsure how to do this please see "Open or close the front access panel door" on page 145.
3. Release the drawbridge mechanism from front access panel door.

Figure 132: Remove the two screws connecting the drawbridge mechanism to the backside of the front access panel door using the T20 T-Shape Key.

Warning
The drawbridge mechanism facet is under tension and will recoil/snap back if not held down while removing the screws.

4. Recoil draw bridge mechanism cord.

Figure 133: With both screws now removed, grab hold of the drawbridge mechanism facet and let the cord recoil (while still attached to the facet), and rest it against the front panel.

5. Unscrew and remove front access panel door.

Figure 134: Unscrew both screws from each back hinge of the front access panel door attached to the front panel using the T25 T-Shape key. When all screws from the hinges are removed, slowly pull the front access panel door away from the unit and set a side.

Warning
SUPPORT THE UNDERSIDE OF THE PANEL WHEN UNSCREWING AS IT WILL OTHERWISE DROP WHEN THE FINAL SCREW IS REMOVED

6. Position and attach replacement door.

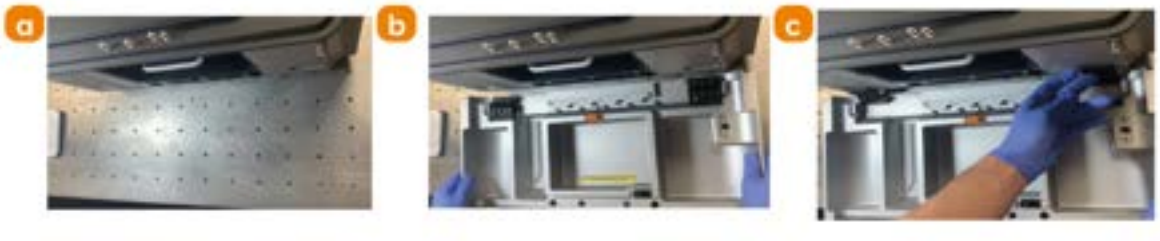


Figure 135: Rest the replacement door face down on the bench top in front of the unit. Turn up the replacement door hinges against the front panel and fine tune the door position such that the hinge through holes line up with the hinge front panel screw holes.

7. Screw in replacement door.

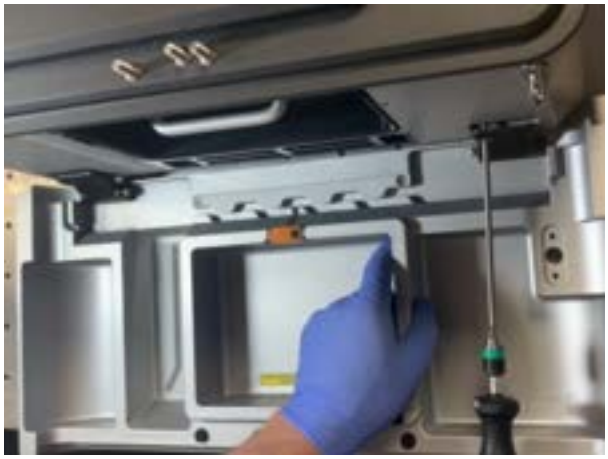


Figure 136: Attach the replacement door to the front panel, screwing two screws through each black hinge using the T25 T-Shape key.

8. Uncoil and pull down drawbridge mechanism cord and facet.

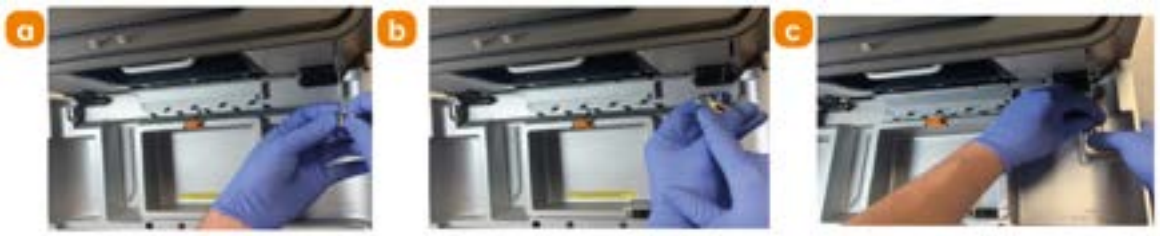


Figure 137: Grab hold of the drawbridge mechanism facet and pull it out to extend the mechanism cord. Extend the cord with enough slack so that it can be positioned on the replacement door.

9. Screw in drawbridge mechanism facet to replacement door.

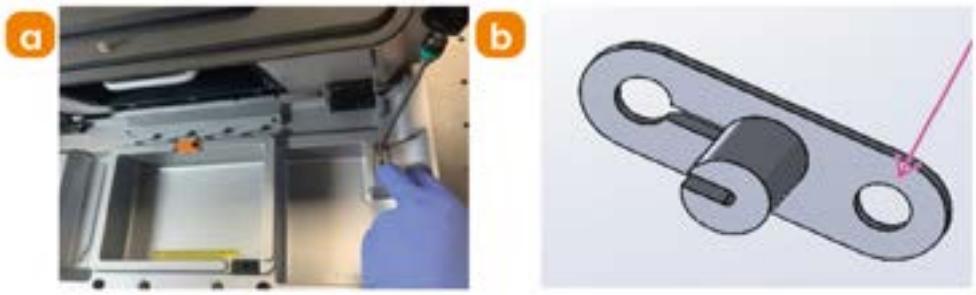


Figure 138: Attach the drawbridge mechanism facet to the backside of the replacement door with two screws using the T20 T-Shape Key. Ensure the circular through-hole of the facet is positioned with the screw hole as shown.

10. Close front access panel door. If you are unsure how to do this please see "Open or close the front access panel door" on page 145.
11. Reconnect the USB cable and power supply and then turn on the instrument. See "Power up Sequence" on page 99
12. Check laser safety interlock.

Figure 139: a) In Fusion, activate the EPI - Green channel. Set the laser power to 50%. Set the exposure time to 100 ms. Click “Live”, and then fully open the top cover lid. b) After 5 minutes of being open, the laser will stop emitting and will not resume until the lid is closed and opened again. c) Observe the blue laser light emitting from the objective lens, and verify that the interlock activates after 5 minutes, is shutting off the laser light completely.

6.18 Front Access Panel Door Cable Latch Replacement

Read before starting procedure	
When should the procedure be performed?	The front access panel door drawbridge mechanism has failed or is broken.
Operation difficulty	Easy
Time to complete	10-15 mins
Replacement part codes	Focus Seek and Lock Module **SV-INS-CBL-MGT-D**
Tools	2.5 mm Hex bit Wera 1/4 in Hex Adjustable Torque Screwdriver, 0.3 →- 1.2 Nm See "Tools List" on page 100.
Laser safety considerations	**The unit must be powered OFF during this procedure.** **If power is re-established while replacing the front access panel door cable latch, Class 2 laser radiation is accessible with the front access panel door and/or the top lid open (but only one laser can be operational), see "Operational laser access. a) Sample holder and b) Focus Seek and Lock opening. " on page 17.** **If the instrument is powered OFF, then there is no hazard.**
Mechanical safety considerations	None

1. Ensure the BC43 unit is powered OFF before starting this procedure.
2. Pull down the front access panel door. If you are unsure how to do this please see "Open or close the front access panel door" on page 145.
3. Unscrew the touch latch using 2.5 mm Hex bit loaded into the AdjTorqGun 0.3 - 1.2 Nm. Screw in the replacement latch using the same tools.

Figure 140: Unscrew the touch latch using 2.5mm Hex bit loaded into the AdjTorqGun 0.3 - 1.2 Nm.

4. Close front access panel door. If you are unsure how to do this please see "Open or close the front access panel door" on page 145.

6.19 Front Access Panel Door Push to Release Mechanism Replacement

Read before starting procedure	
When should the procedure be performed?	The front access panel door drawbridge mechanism has failed or is broken.
Operation difficulty	Easy
Time to complete	10 - 15 mins
Replacement part codes	Front Access Panel Door **SV-INS-LAT**
Tools	T8 Key See "Tools List" on page 100.
Laser safety considerations	**The unit must be powered OFF during this procedure.** **If power is re-established while replacing the push to release mechanism, Class 2 laser radiation is accessible with the front access panel door and/or the top lid open (but only one laser can be operational), see "Operational laser access. a) Sample holder and b) Focus Seek and Lock opening. " on page 17.** **If the instrument is powered OFF, then there is no hazard.**
Mechanical safety considerations	None

1. Ensure the BC43 unit is powered OFF before starting this procedure.
2. Pull down front access panel door. If you are unsure how to do this please see "Open or close the front access panel door" on page 145.
3. Unscrew the 4 push-to-release socket mechanism screws using the T8 key. Screw in the replacement using the same tool.

Figure 141: Unscrew the push-to-release socket mechanism.

4. Unscrew the plug of the push-to-release mechanism using the T8 Key. Screw in the replacement using the same tool.

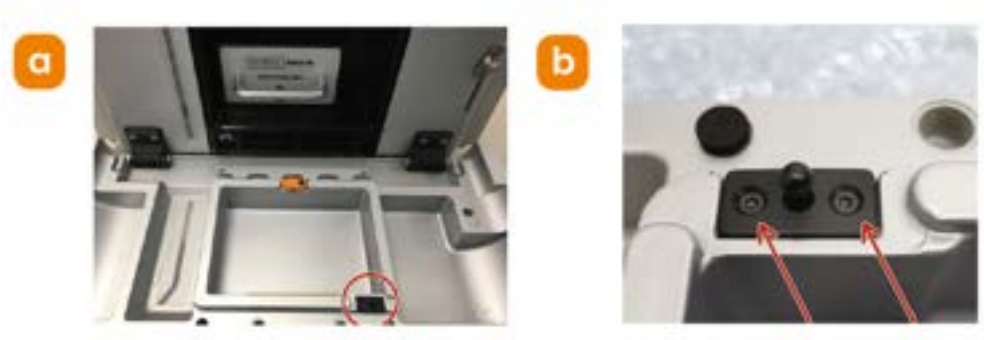


Figure 142: Push-to-release plug mechanism a) placement and b) screw positions.

5. Close front access panel door. If you are unsure how to do this please see "Open or close the front access panel door" on page 145.

6.20 Returning the BC43 for Maintenance

To return the BC43 for maintenance or repair please contact our customer support team who will be able to advise you further.

Section 7 - Advanced Maintenance Procedures

This section of the user guide covers advanced maintenance procedures provided as part of BC43 Install Qualification service package, please contact your local support representative for more information or to purchase this service.

The following advanced maintenance procedures are designed to be performed as part of the IQ service package. Thereafter, the material in this guide is designed to be used as a reference when required, recommended intervals between procedure occurrences are listed in the table below.

Table 2: Recommended interval times between repeating procedures.

Procedure	Procedure Recurrence Interval
"Montage Functionality Check Using a Standard Sample " on page 182	**Every 3 months**
"Z-scan Functionality Check Using a Standard Sample" on page 186	**Every 3 months**
"Multichannel Functionality Check Using a Standard Sample" on the next page	**Every 3 months**
"Transmitted Light Imaging Functionality Check Using a Standard Sample" on page 171	**Every 3 months**
"Laser Throughput Check Using a Calibrated Sample" on page 191	**Maximum interval 3 months**

In the course of performing these qualitative measurements, as part of the IQ service package, acquired images will need to be saved for future comparisons of system performance. We recommend that images are saved at the following location C:\Program Files\Fusion\Support.

Copies of protocol files and an excel sheet to calculate and record system laser powers are available from our Downloads area.

7.1 Multichannel Functionality Check Using a Standard Sample

Read before starting procedure	
When should the procedure be performed?	These instructions describe a simple method to check and verify the proper activation of the BC43 fluorescence channels (lasers and emission filter wheel) by imaging a standard multi-labeled sample which has a known fluorescence response when viewed through each channel.
Operation difficulty	Easy
Time to complete	15 minutes
Tools	BC43 Install Qualification slide pack (XS-SMPL-TEST-SET) For this procedure you will need the protocol file **BC43 Confocal Multichannel Functionality Check ENI-02263.bkp** Download protocol files from our Downloads area
Laser safety considerations	**Class 2 laser radiation is accessible with the lid open at the microscope objective lens or objective lens port, but only one laser can be operational, see "Operational laser access. a) Sample holder and b) Focus Seek and Lock opening. " on page 17.**
Mechanical safety considerations	None

1. Power ON the BC43 instrument. See "Power up Sequence" on page 99 for further guidance.
2. Run Fusion software.
3. Adjust Fusion image naming preferences. In the preferences pane, click the file manager button. For this functionality check, it is recommended that the image filename be set using the Date, Time, and Protocol options in this order so that the saved image(s) can easily be retrieved in the future. See File manager in the Fusion online help for more information on these settings. Click the **Back to Imaging** button once these image file naming preferences have been adjusted.
4. Load the GATTA-Cells 4C standard sample into the stage insert (coverslip facing down toward the objective lens nosepiece). Please see "Add a Sample" on page 71 and "Inserting/Removing the Sample Holder" on page 101 for further guidance on adding or removing a sample.

5. In Fusion, engage a low magnification objective lens (other than 2x). Click the "**Find Coverslip**" button.
6. When focus has been achieved, switch to a high magnification objective lens.

Note
This test is intended to be used with the 40x, 0.75 NA objective lens if present, but other objective lenses can be used as well.

7. Turn on the image automapping option to facilitate visualization of the different fluorescent images of the sample that will be acquired in the next few steps.

Figure 143: If not already on, turn ON the image automapping feature.

8. Download the **BC43 Confocal Multichannel Functionality Check ENI-02263.bkp** Fusion protocol to the PC workstation.
9. In the Fusion Protocol Manager, Click "**Import**" and open the Installation Quality Multi-channel protocol from the indicated Windows file directory. Click "**Overwrite**" when the "**Imported channel(s) already exists**" message prompt appears.
10. Prepare acquisition settings.

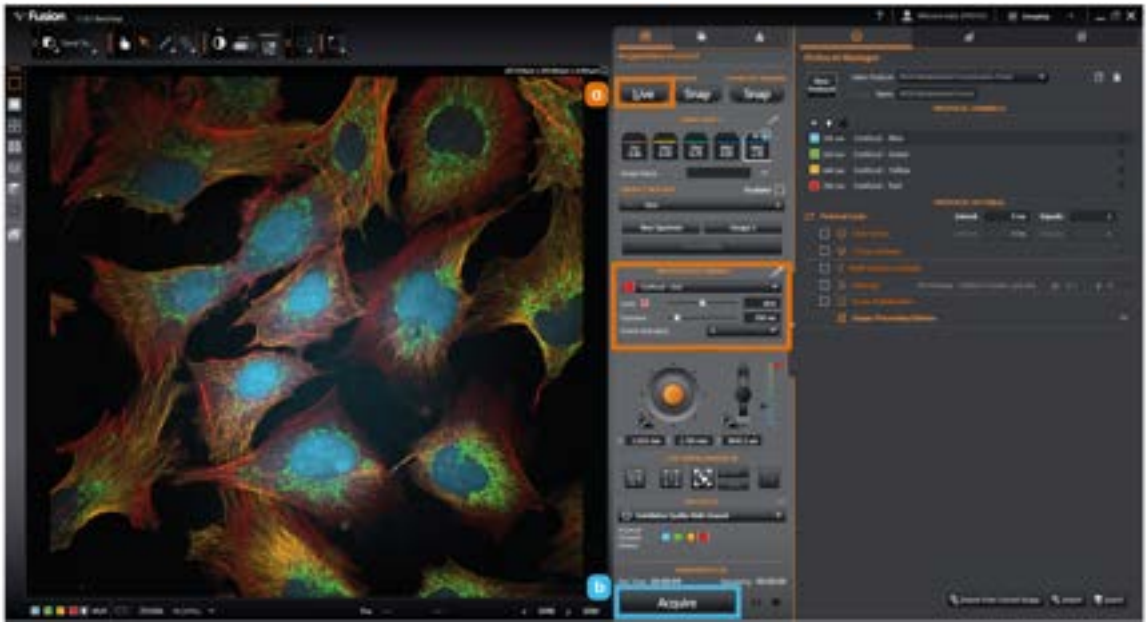


Figure 144: a) Click "Live" and adjust the focus to midplane of the cellular specimen. Successively cycle through the 4 confocal channels (Blue, Green, Yellow, Red) and adjust the laser powers if necessary to achieve a good signal-to-noise ratio for each channel image. b) Then Click "Acquire" in the Protocol Manager to capture a 4-channel fluorescence image of the specimen.

11. Choose the 2D split channel mode view  of the sample.

Note
The 4-channel image should clearly exhibit the following morphological characteristics with little to no signs of channel bleed-through or cross-talk: Confocal -Blue: nuclei (DAPI); Confocal - Green: mitochondria (Alexa Fluor 488); Confocal - Yellow: microtubules (Alexa Fluor 555); Confocal - Actin (Alexa Fluor 647).

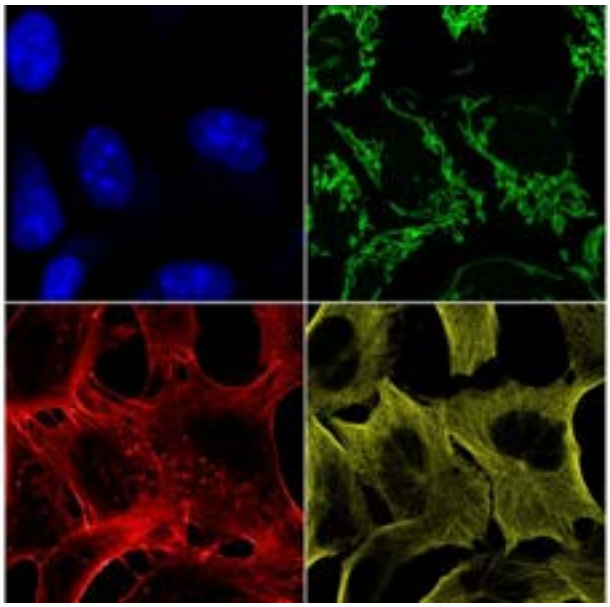


Figure 145: Sample images

12. In Windows, navigate to the directory that has been designated the Fusion Root Folder. Copy the raw .ims and metadata image files created during this functionality check and paste them into the C:\Program Files\Fusion\Support folder directory. Image files will be required for future comparisions of system performance.
13. Remember to reset the image file naming preferences back to your preferred settings after the functionality check has been completed.

7.2 Transmitted Light Imaging Functionality Check Using a Standard Sample

Read before starting procedure	
When should the procedure be performed?	These instructions describe a simple method to diagnose and verify the functionality of the transmitted light imaging modes - Brightfield and Differential Phase Contrast (DPC) - of the microscope using a standard sample. Proper activation of the transmitted light LED array turret is checked first, and then brightfield and DPC images of a prepared slide of diatoms are inspected for expected structural features which appear clearly only when these instrument image modes are operating correctly. Note: A high numerical aperture oil immersion objective lens is required for this procedure.
Operation difficulty	Very easy
Time to complete	15 minutes
Tools	BC43 Install Qualification slide pack (XS-SMPL-TEST-SET) Cleaning swabs Immersion oil See "Tools List" on page 100
Laser safety considerations	**Class 2 laser radiation is accessible with the lid open at the microscope objective lens or objective lens port, but only one laser can be operational, see "Operational laser access. a) Sample holder and b) Focus Seek and Lock opening. " on page 17.**
Mechanical safety considerations	None

1. Power ON the BC43 instrument. See "Power up Sequence" on page 99 for further guidance.
2. Run Fusion software.
3. Verify LED turret illumination using Brightfield channel. Lift open the top cover lid of the instrument.

Figure 146: In the Fusion remote panel, under the Navigation Channel heading, activate the Brightfield channel. Set the transmitted LED turret Intensity to 100%. Set the Exposure time to 10 ms.

4. Now click the Live button in the Fusion remote panel to activate a live preview and turn on the transmitted light turret.

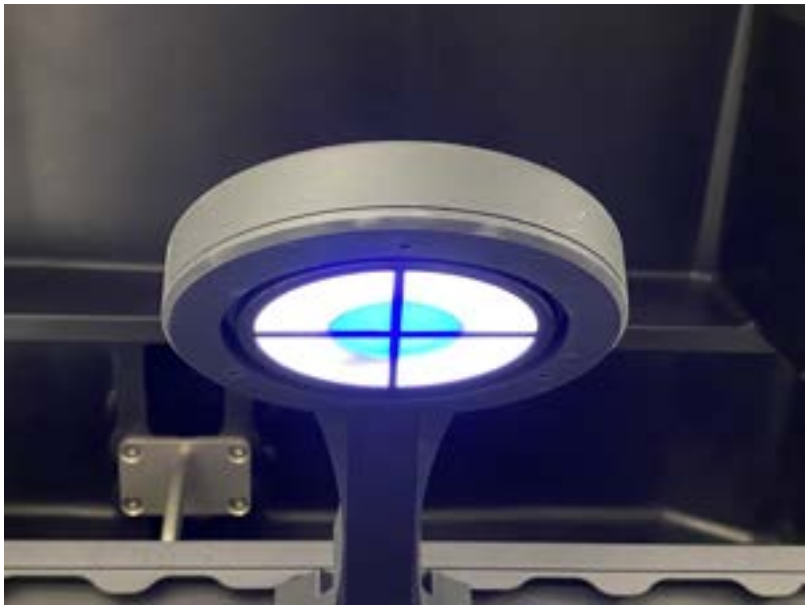


Figure 147: With the top cover lid open, squat down and observe the underside of the transmitted light turret from a distance at an oblique angle. Verify that outer annulus of the LED turret is constantly ON and has a white colour as shown. Also verify that the central section of the LED turret is constantly ON and exhibits a blue colour as shown.

5. Verify LED turret illumination using DPC High Speed channel

Figure 148: In the Fusion remote panel, under the Navigation Channel heading, activate the Differential Phase Contrast channel. Choose the High Speed mode. Set the transmitted LED turret Intensity to 10%. Set the Exposure time to 10 ms.

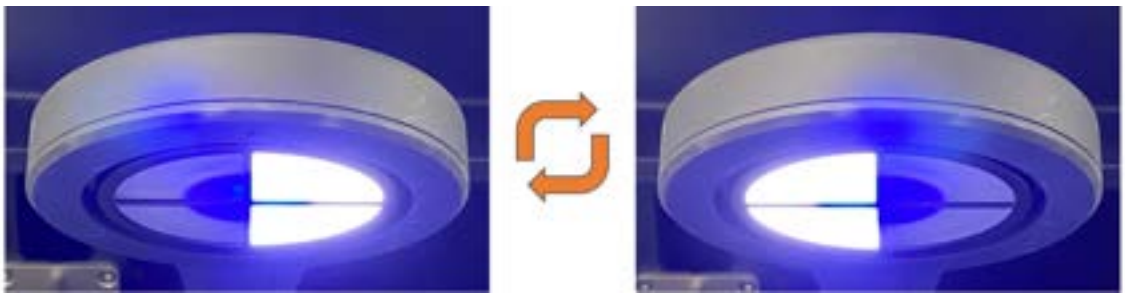


Figure 149: With the top cover lid open, squat down and observe the underside of the transmitted light turret from a distance at an oblique angle again. Verify that LED turret rapidly alternates turning ON/OFF the left/right halves of the turret.

6. Verify LED turret illumination using DPC High Quality channel.

Figure 150: In the Fusion remote panel, under the Navigation Channel heading, activate the Differential Phase Contrast channel. Choose the High Quality mode. Set the transmitted LED turret Intensity to 10%. Set the Exposure time to 10 ms.

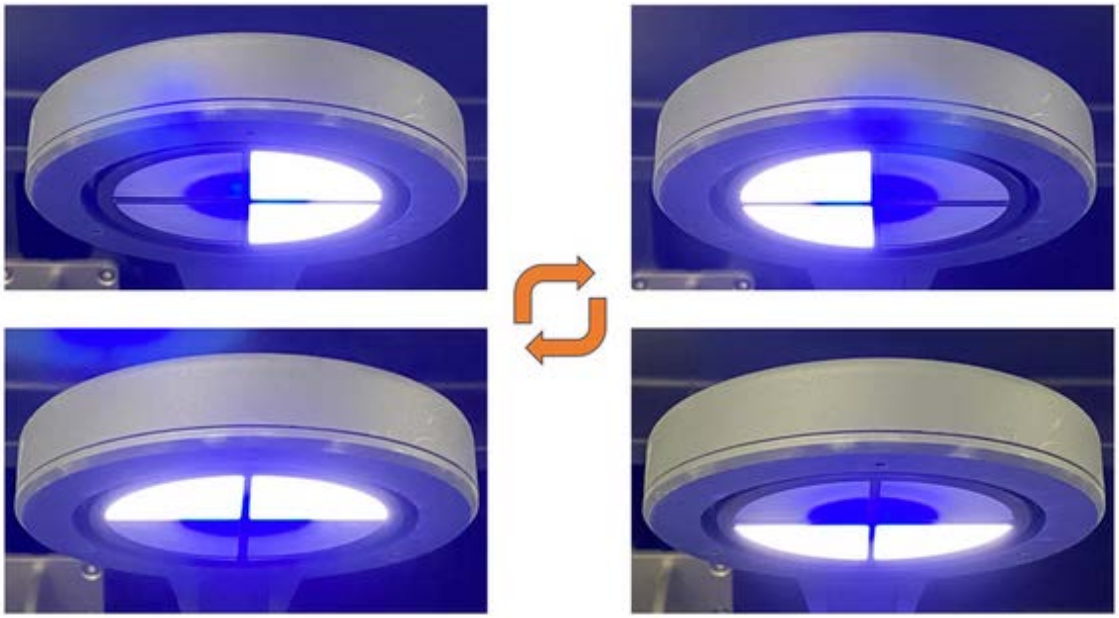


Figure 151: With the top cover lid open, squat down and observe the underside of the transmitted light turret from a distance at an oblique angle again. Verify that LED turret rapidly alternates turning ON/OFF the left/right/top/bottom halves of the turret.

7. Clean Diatom Test Slide

Figure 152: Apply a few drops of optical grade lens cleaning solvent to tip of a cleaning swab. Thoroughly clean the coverslip surface and the backside of the slide within the black encircled area of the sample using an outwardly spiralling motion of the swab in contact with the glass surfaces.

8. Mount Diatom Test Slide into sample holder. Please see "Add a Sample" on page 71 and "Inserting/Removing the Sample Holder" on page 101 for further guidance on adding or removing a sample.

9. Translate XY-stage to align Diatom Test Slide to objective lens

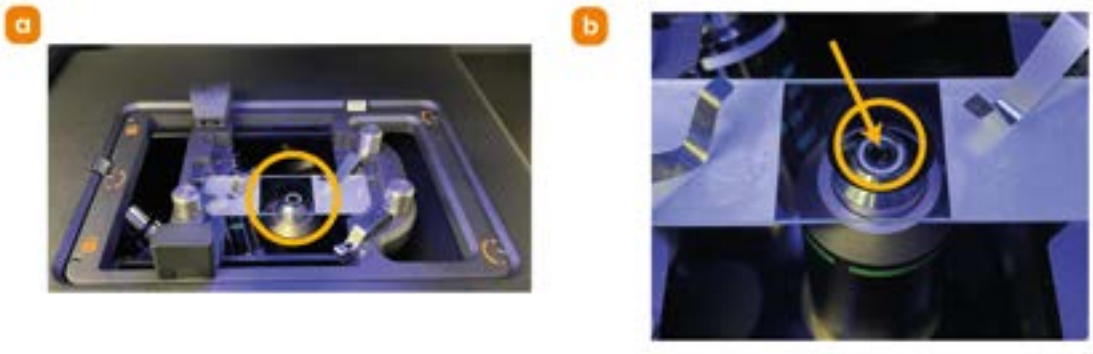


Figure 153: After the Diatom Test slide has been properly mounted into the sample holder, translate the XY-stage until the small black circled area within the Diatom Test slide is positioned over the active objective lens.

10. Now close the top cover lid of the BC43 unit.
11. Autofocus on diatoms using air objective lens

Figure 154: a) In the Fusion remote panel, under the Navigation Channel heading, select the Brightfield channel and set the Intensity to 10% and the Exposure time to 50 ms. b) Activate a low magnification objective lens compatible with the Find Coverslip software feature such as a 20x air objective. c) Click the Live button to activate the live preview. d) Click the Find Coverslip button to automatically find focus on the test slide coverslip.

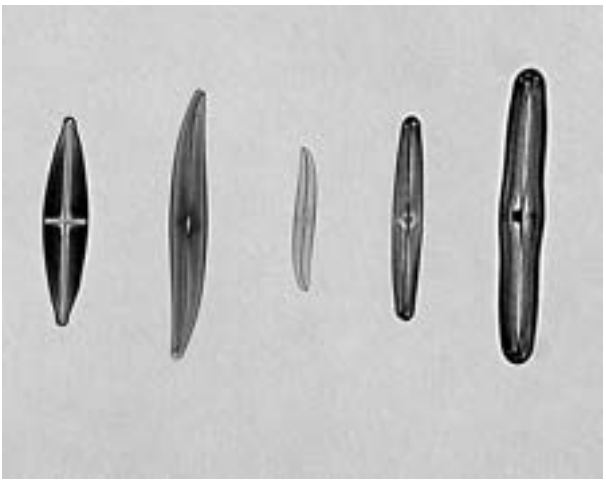


Figure 155: Adjust the focus and translate the XY-stage to bring into view and center the 5 diatoms in the test sample within the camera field of view.

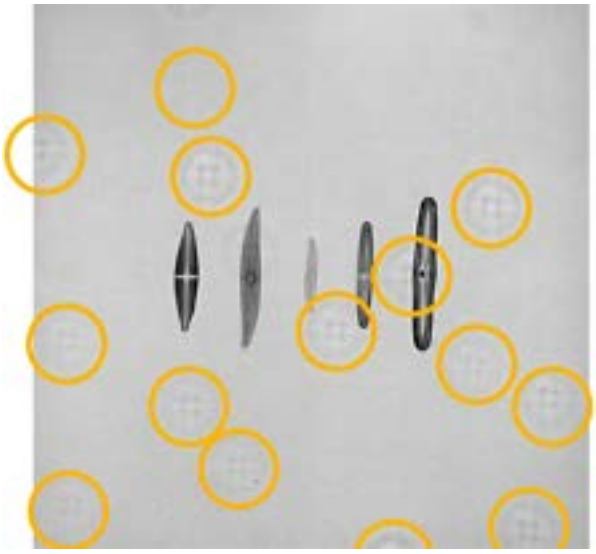


Figure 156: If any circular-shaped, out-of-focus cross patterns are present in the background of the image, remove the test slide from the stage insert and clean both sides of the slide again as previously described.

Note
If any background artefacts as shown in the image above persist even after cleaning the slide again, then clean all other accessible optical surfaces. **See** "Cleaning the Objective Lenses" on page 130 **See** "Cleaning the Infinity Space Window" on page 132 **See** "Cleaning the Focus Seek and Lock Module" on page 134

12. Switch to 60x oil immersion objective lens. A confirmation dialog box will appear warning about the transition to an oil objective lens. Click Yes to confirm.

    Once the objective has moved to the oiling position, add a drop of immersion oil to the top lens of the objective. After oil has been applied to the 60x immersion objective lens, click Complete in the Adding Oil dialog box and wait for the XY-stage to move back to the sample position and the immersion objective lens to refocus onto the coverslip interface. If you are unfamiliar with adding oil to the oil immersion objective please see "Add/Remove Immersion Oil to the Oil Immersion Objective Lens" on page 119 for full instructions.

Figure 157: a) Switch to the oil immersion objective. b) change the Exposure time for the Brightfield channel to 50 ms and verify the light source Intensity is still set to 50%.

Figure 158: If not already on, turn ON the image automapping feature to facilitate visualization of the diatoms and this magnification.

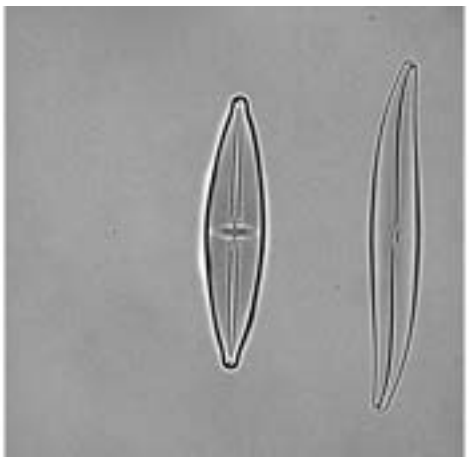


Figure 159: Finely adjust the focus again, and translate the XY-stage to center the left-most diatom (Stauroneis phoenicenteron) into the field of view. Shown on left above.

13. Examine and count striae in Stauroneis phoenicenteron diatom using Brightfield channel. Snap and save a Brighfield channel image of the Stauroneis phoenicenteron diatom centered in the field of view.

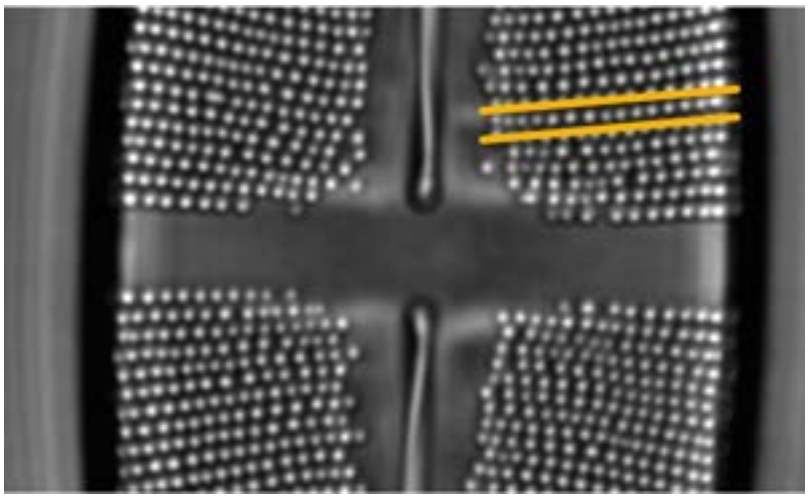


Figure 160: Hover over the image with the mouse pointer and use the scroll wheel to digitally zoom the saved image. 15-18 small, round and distinct striae structures should be clearly visible and countable in a row radiating outwards from the central region of the diatom.

14. If necessary, finely re-adjust the objective lens focus in 100 nm steps up or down during a live preview until the striae are as sharply visible on the monitor

screen as possible.

15. Examine and count striae in Stauroneis phoenicteron diatom using Differential Phase Contrast High Quality channel

Figure 161: Change the Navigation Channel to the Differential Phase Contrast channel. Set the Exposure time to 50 ms and choose the High Quality Mode.

16. Click the Live button to activate another live preview and once again finely adjust the focus in 100 nm z-steps until the striae of the diatom are visible again, this time showing a shadowed relief appearance. Snap and save an image of the diatom in this channel once optimized focus has been achieved.

17. As before, 15-18 individual striae should be visible and countable, matching the same number in the row that was examined in step 13.

18. In Windows, navigate to the directory that has been designated the Fusion Root Folder. Copy the raw .ims and metadata image files created during this functionality check and paste them into the C:\Program Files\Fusion\Support folder directory. Image files will be required for future

comparisions of system performance.

7.3 Montage Functionality Check Using a Standard Sample

Read before starting procedure	
When should the procedure be performed?	These instructions provide a simple method to check and verify the proper operation of the motorized BC43 XY-stage by running a montage protocol on a standard specimen that spans several fields of view at high magnification and readily shows visible discontinuities if stage motion errors occur.
Operation difficulty	Very Easy
Time to complete	15 Minutes
Tools	Convallaria Montage Sample Slide (XS-MONT-SMPL) For this procedure you will need the protocol file **BC43 Montage Functionality Check ENI-02262.bkp** Download protocol files from our Downloads area
Laser safety considerations	**Class 2 laser radiation is accessible with the lid open at the microscope objective lens or objective lens port, but only one laser can be operational, see "Operational laser access. a) Sample holder and b) Focus Seek and Lock opening. " on page 17.**
Mechanical safety considerations	None

1. Power ON the BC43 instrument. See "Power up Sequence" on page 99 for further guidance.
2. Run Fusion software.
3. Adjust Fusion image naming preferences. In the preferences pane, click the file manager button. For this functionality check, it is recommended that the image filename be set using the Date, Time, and Protocol options in this order so that the saved image(s) can easily be retrieved in the future. See File manager in the Fusion online help for more information on these settings. Click the **Back to Imaging** button once these image file naming preferences have been adjusted.
4. Load the standard **Montage sample (Convallaria)** into the sample holder (coverslip facing down toward the objective lens nosepiece). Please see "Add a Sample" on page 71 and "Inserting/Removing the Sample Holder" on page 101 for further guidance on adding or removing a sample.

5. In Fusion, engage a low magnification air objective lens (other than 2x). Click the "**Find Coverslip**" button.
6. When focus has been achieved, switch to a high magnification objective lens.

Note
This test is intended to be used with the 40x, 0.75 NA objective lens if present, but other objective lenses can be used as well.

7. Turn on the image automapping option to facilitate visualization of the different fluorescent images of the sample that will be acquired in the next few steps.

Figure 162: If not already on, turn ON the image automapping feature.

8. Download the BC43 Montage Functionality Check ENI-02262.bkp Fusion protocol to the PC workstation. In the Fusion Protocol Manager, Click "**Import**" and open the **BC43 Montage Functionality Check ENI-02262.bkp** protocol from the indicated Windows file directory.
9. Click "Overwrite" when the "Imported channel(s) already exists" message prompt appears.
10. Click Live and finely adjust the focus to midplane of the cellular specimen using the Confocal - Green channel.

Figure 163: a) Click Live and finely adjust the focus to midplane of the cellular specimen using the Confocal - Green channel. b) Once the protocol is imported navigate to the Image Processing Option area and ensure Stitching is set to ON. c) Click "Acquire" in the Protocol Manager to capture a 8x8 stitched montage of the specimen.

Note
If using different objective lens other than 40x, the protocol montage height and width should be adjusted to fully capture the entire Convallaria cross-section.

11. Closely examine the result of the stitched Convallaria image. Zoom in to the image and scan around to see if there are any obvious regions where the stitching algorithm has failed to properly connect the image tiles.

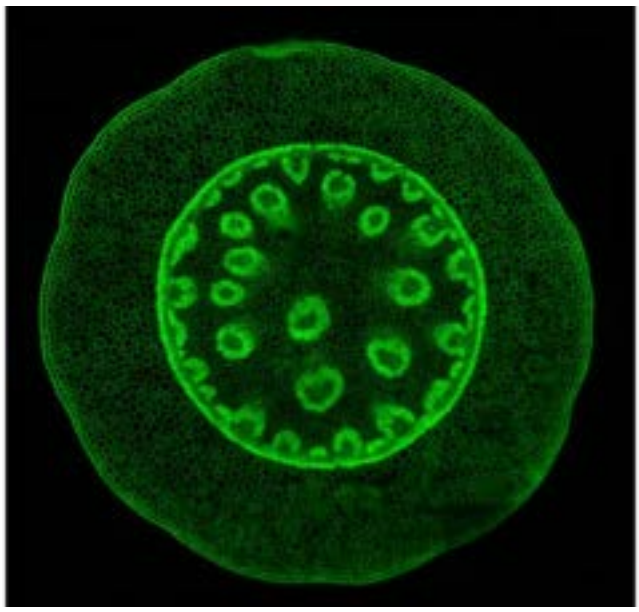


Figure 164: Convallaria sample image

12. In Windows, navigate to the directory that has been designated the Fusion Root Folder. Copy the raw .ims and metadata image files created during this functionality check and paste them into the C:\Program Files\Fusion\Support folder directory. Image files will be required for future comparisions of system performance.

13. Remember to reset the image file naming preferences back to your preferred settings after the functionality check has been completed.

7.4 Z-scan Functionality Check Using a Standard Sample

Read before starting procedure	
When should the procedure be performed?	These instructions detail a simple method to test and verify the ability of the instrument to properly step the Z-stage during an axial scan (Z-scan) of a thick standard specimen and create an accurate 3D representation of its tubular, filamentous, and spherical microstructures thereafter.
Operation difficulty	Very Easy
Time to complete	15 mins
Tools	BC43 Install Qualification slide pack (XS-SMPL-TEST-SET) For this procedure you will need the protocol file **BC43 Z-scan Functionality Check ENI-02261.bkp** Download protocol files from our Downloads area
Laser safety considerations	**Class 2 laser radiation is accessible with the lid open at the microscope objective lens or objective lens port, but only one laser can be operational, see "Operational laser access. a) Sample holder and b) Focus Seek and Lock opening. " on page 17.**
Mechanical safety considerations	None

1. Power ON the BC43 instrument. See "Power up Sequence" on page 99 for further guidance.
2. Run Fusion software.
3. Load the Aspergillus Z-scan Sample Slide onto the sample holder (coverslip facing down toward the objective lens nosepiece). Please see "Add a Sample" on page 71 and "Inserting/Removing the Sample Holder" on page 101 for further guidance on adding or removing a sample.
4. Translate the XY-stage using the 3D Connexion joystick or the software joystick to position the blue-colored fungus strip over the active objective lens.
5. In Fusion, engage a low-power air objective lens (**10x, 20x, or 40x**). Click the "**Find Coverslip**" button.

6. Turn on the image automapping option to facilitate visualization of the different fluorescent images of the sample that will be acquired in the next few steps.

Figure 165: If not already on, turn ON the image automapping feature.

7. Adjust Fusion image naming preferences. In the preferences pane, click the file manager button. For this functionality check, it is recommended that the image filename be set using the Date, Time, and Protocol options in this order so that the saved image(s) can easily be retrieved in the future. See File manager in the Fusion online help for more information on these settings. Click the **Back to Imaging** button once these image file naming preferences have been adjusted.

8. When focus has been achieved, switch to a high magnification objective lens.

Note
This test is intended to be used with the 40x, 0.75 NA objective lens if present, but other objective lenses can be used as well.

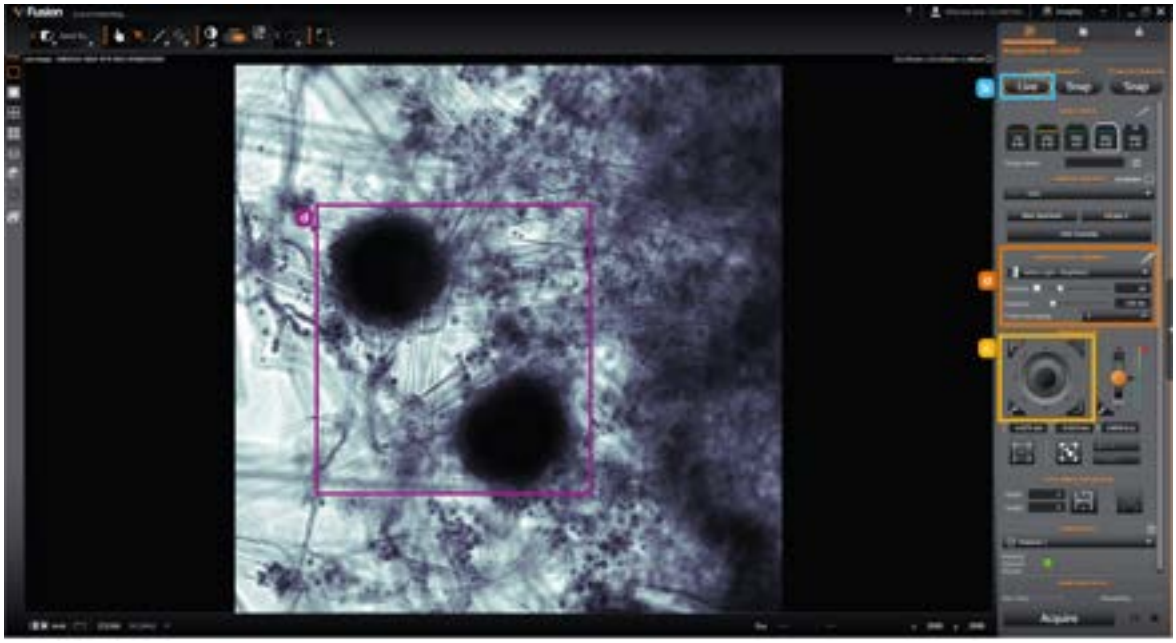


Figure 166: a) Activate the Brightfield channel with an light Intensity of 20% and Exposure time of 150 ms. b) Click Live to activate a live preview of the sample. c) Translate the XY-stage to a region of the sample that contains at least one round conidia spore of the fungus. d) In the Brightfield transmitted light channel, the spore will appear as a large, dark, circular feature.

9. Download the **BC43 Z-scan Functionality Check ENI-02261.bkp** Fusion protocol to the PC workstation.

10. In the Fusion Protocol Manager, Click "**Import**" and open the Installation Quality Z Scan protocol from the indicated Windows file directory. Click "**Overwrite**" when the "**Imported channel(s) already exists**" message prompt appears.

Figure 167: a) Activate the Confocal - Red channel, then select Live. Translate the XY stage to a mid-plane of the Aspergillus sample that contains at least one round conidia spore of the fungus. Adjust the laser power again if necessary. Check the preview image using the Confocal - Yellow channel as well and adjust the laser power again if necessary .b) Finely adjust the focus to midplane of one of the fungal spores and then, under the Z scan settings of the protocol, click the Set button of the Scan Centre option. c) Click "Acquire" to capture 75 µm Z-scan (500 nm z-steps) of the specimen at the chosen magnification using both the Confocal - Yellow and Confocal - Red channels.

11. Click "**Acquire**" to capture 75 µm Z-scan (500 nm z-steps) of the specimen at the chosen magnification using both the Confocal - Yellow and Confocal - Red channels.

12. When the Z scan protocol has completed, click on Z-production render option  .

13. Click on the 3D Maximum Intensity Volume view  of the active image.

Figure 168: a) Click on the 3D Maximum Intensity Volume view. b) With the mouse, left-click and grab hold of the 3D rendered image of the fungus and rotate it around in 3D space. Closely examine the result of 3D reconstruction move around to see if there are any obvious regions where the rendering has failed to display properly.

Note
While hovering over any section of the image with the mouse, use the mouse scroll wheel to zoom in or out on the rendering.

14. Closely examine the result of the 3D reconstruction and move it around to see if there are any obvious regions where the Z-scan has not properly captured the spherical shape of the spore or where the rendering has failed to display properly.
15. In Windows, navigate to the directory that has been designated the Fusion Root Folder. Copy the raw .ims and metadata image files created during this functionality check and paste them into the C:\Program Files\Fusion\Support folder directory. Image files will be required for future comparisions of system performance.
16. Remember to reset the image file naming preferences back to your preferred settings after the functionality check has been completed.

7.5 Laser Throughput Check Using a Calibrated Sample

Read before starting procedure	
When should the procedure be performed?	This guide contains a set of instructions that describe how to use a set of fluorescent plastic slides to indirectly infer the laser power for each channel reaching the back of the objective lens. Each plastic slide has been calibrated for their fluorescence intensity response on a ***specific Benchtop Microscope identified by its serial number*** under certain image acquisition conditions. Therefore, under identical acquisition settings, the camera signal can be used to inversely calculate the laser power at any later time. The calibrated plastic slides **MUST** be used **ONLY** with the **Benchtop Microscope they were calibrated on**. Use of the plastic slides with a different Benchtop Microscope unit will yield erroneous power measurements. Contact your local Oxford Instruments - Andor service representative if the calibrated plastic slides received with your Benchtop Microscope are lost or damaged. The indirect measurement of laser power by this method is accurate to within 5% the calculated values, and so it can only be used to confidently deduce power changes that exceed this amount. The advantage of this approach, however, is that it can easily and safely assess considerable laser power variations remotely without the use of a traditional laser power meter or direct access to the laser beam.
Operation difficulty	Very Easy
Time to complete	30 mins
Tools	BC43 Install Qualification slide pack (XS-SMPL-TEST-SET) Download the spreadsheet to calculate and record the power measurements of the system from our Downloads area.
Laser safety considerations	**Class 2 laser radiation is accessible with the lid open at the microscope objective lens or objective lens port, but only one laser can be operational, see "Operational laser access. a) Sample holder and b) Focus Seek and Lock opening. " on page 17.**
Mechanical safety considerations	None

1. Retrieve the **XS-SMPL-TEST-SET** box of Calibration Slides and Standard Samples that were received with the microscope. Confirm the Unit Serial

Number is written on the bottom of this box matches the Unit Serial Number of the microscope that is being tested for laser power.

2. Power ON the BC43 instrument. See "Power up Sequence" on page 99 for further guidance.
3. Run Fusion software.
4. Adjust Fusion image naming preferences. In the preferences pane, click the file manager button. For this functionality check, it is recommended that the image filename be set using the Date, Time, and Protocol options in this order so that the saved image(s) can easily be retrieved in the future. See File manager in the Fusion online help for more information on these settings. Click the **Back to Imaging** button once these image file naming preferences have been adjusted.
5. Load calibrated green fluorescent plastic slide into the sample holder.

Figure 169: a) Obtain the calibrated green fluorescent plastic slide. b) Note the full serial number code associated with this slide of the format PWR-CAL-SLD-####, where #### is a 4-digit number. Also note the calibration constant, K, written on the slide label. c) Lift open the top cover lid of microscope, load the green plastic slide into the sample holder as shown, and then close the top cover lid.

6. Locate slide edge.

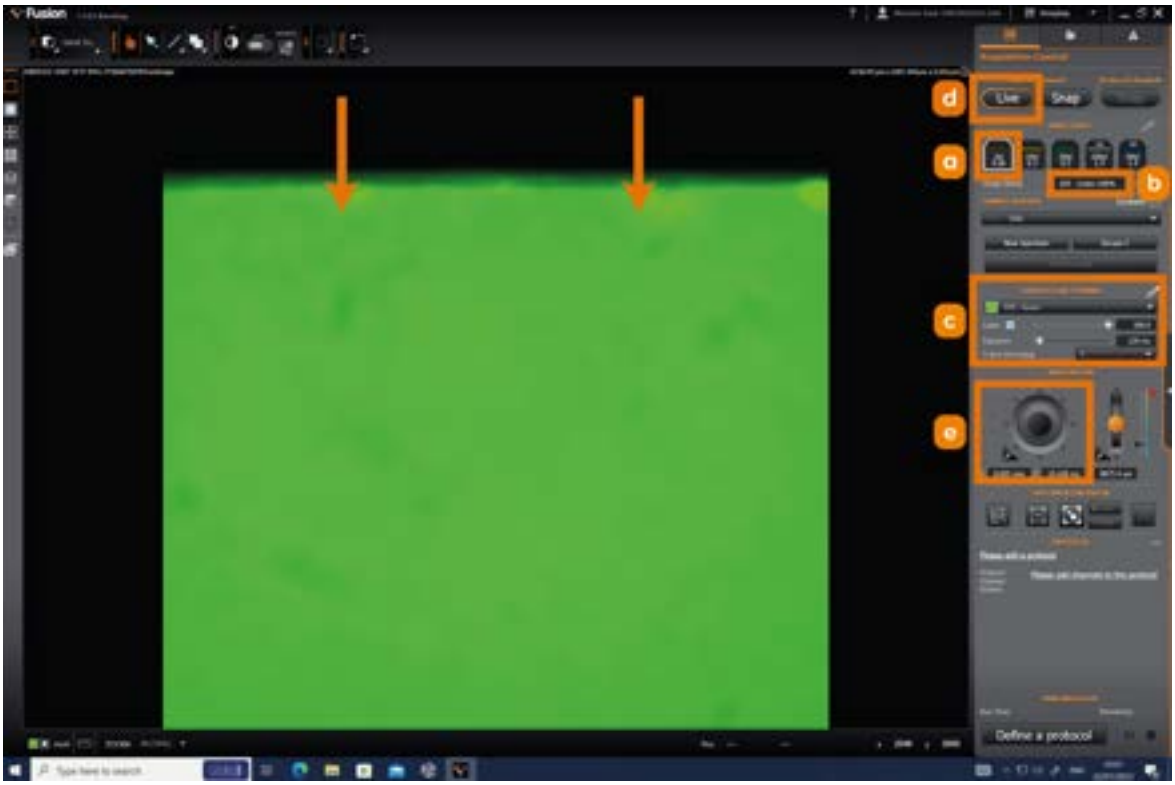


Figure 170: a) In the Fusion remote panel, select and activate the 2x objective lens. b) Set the image name to EPI-Green 100%. c) Select and activate the EPI-Green channel. Set the 488 nm laser power to 100%. Set the exposure time to 120 ms. Set the camera Frame Averaging option to1 if it isn't already. The acquisition exposure time will be fixed in a later step. Choose an exposure time that provides enough signal to see the specimen and observe real-time movements of the XY-stage and Z-focus drive. d) Click the Live button to activate a live preview of the sample. e) Laterally translate the specimen using the software or 3D Connexion joystick until the unfocused top edge of the green plastic slide is located.

7. Focus onto slide edge.

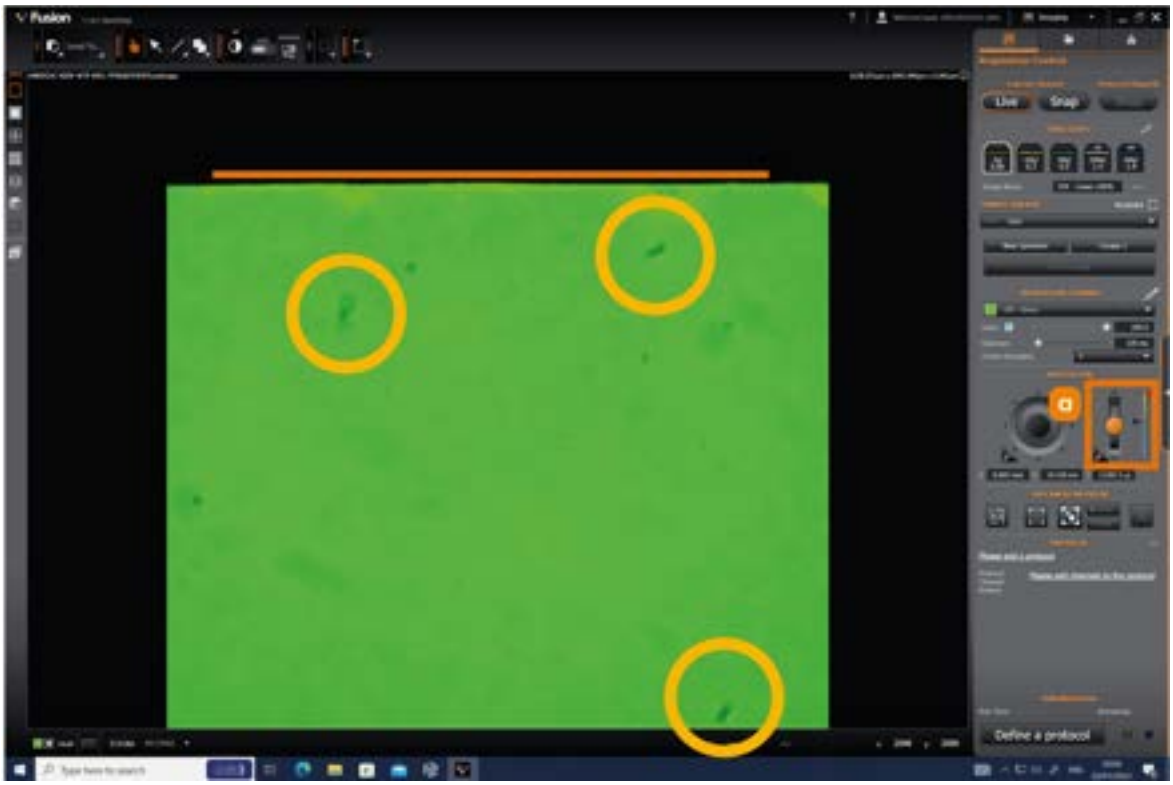


Figure 171: a) Using the software focus lever or the 3D Connexion joystick, adjust the focus position to bring the slide edge and surface defects/imperfections of the slide into sharp focus.

8. Translate slide to marked position (without changing the current Z-focus position). Lift the lid and using the software joystick or the 3D Connexion joystick translate the slide such that the illuminated square of light is between the four dots marked on the slide.

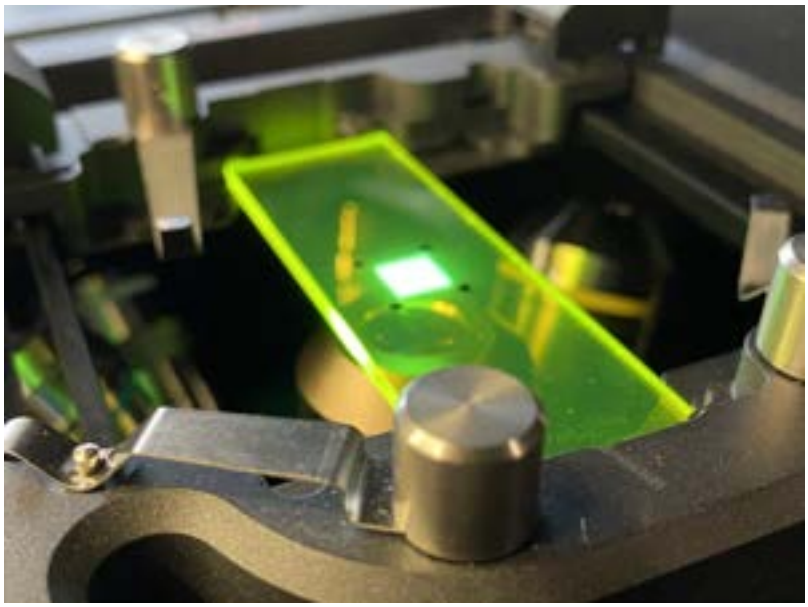


Figure 172: Translate the slide such that the illuminated square of light is between the four dots marked on the slide.

9. Adjust exposure time for maximum signal without excessive pixel saturation.

Figure 173: a) Open Fusion's expander panel by clicking on the indicated button on the side of the remote panel. b) Click on the Image Analysis tab in the expander panel. c) Click the Show Regions Histogram button and then turn ON the Display using Logarithmic scale button. d) In the Image Viewer Panel, click on the Enhance Contrast button to open the live image histogram. Click the Display using Logarithmic scale box. Change the image histogram Min value to 100. Change the image histogram Max value to 65535. e) Now increase or decrease the exposure time in increments of +/- 10 ms until a maximum average image signal is achieved across the entire field of view without an excessive number of pixels reaching the saturation value of 65535 (<100 is ideal). Note the chosen exposure time that satisfies this condition.

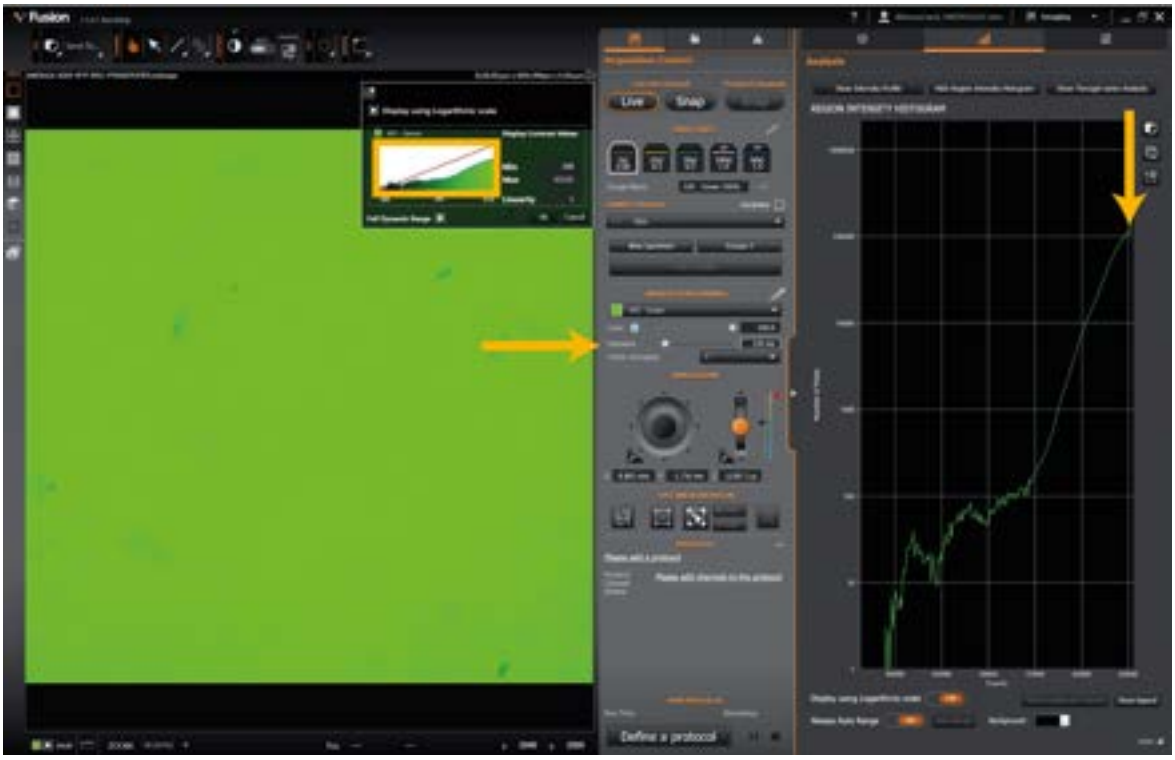


Figure 174: Above demonstrates where the exposure time has been increased too high (150 ms) leading to excessive pixel saturation over many image pixels.

10. Once an Exposure time that minimizes pixel saturation has been settled on, note this exposure time and then stop the live preview.

Note
It is important that you STOP the live preview first before clicking the Snap button in the next instruction!

11. Acquire and save EPI - Green 100% image and measure mean pixel intensity.

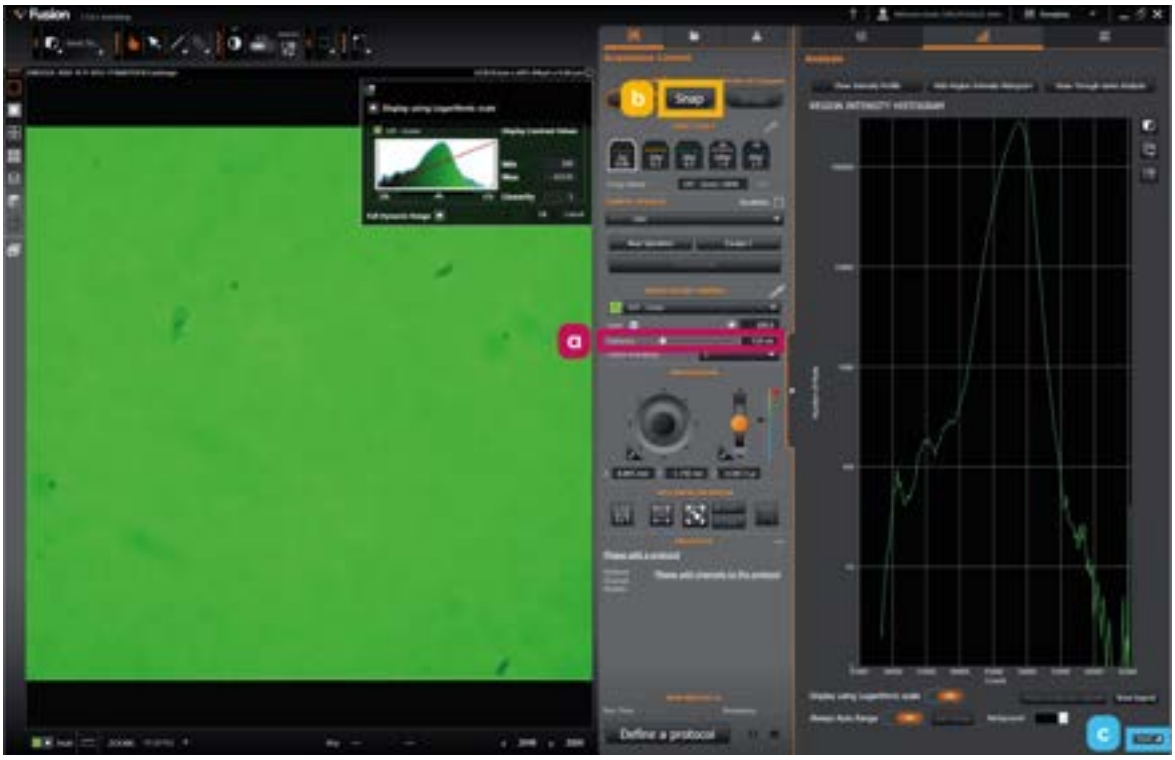


Figure 175: a) Make a note of the exposure time that minimizes pixel saturation. b) Click the Snap button to capture and save the EPI-Green 100% laser power image of the Green laser throughput calibration slide. c) In Fusion's Expander Panel, under the Image Analysis tab, click the More option at the bottom right of the image histogram.

Figure 176: Note the mean pixel intensity value listed under the Region Intensity Histogram Statistics (48492 in this example).

12. An Excel sheet to calculate and record laser powers is available to download, if you have already downloaded protocol files for other procedures a copy of the Excel file is in the zip file associated with this download.

    Calculate EPI - Green channel 488 nm laser power at objective lens.

$$p_{eg} = \frac{int_{eg}}{(t_{eg} \times K)}$$

    p_{eg} - laser power at objective lens for confocal green channel

    int_{eg} - measured mean pixel intensity of confocal green channel

    t_{eg} - exposure time of confocal green channel

    K - calibration constant of slide

    The uncertainty associated with this laser power is +/- 5% the calculated value.

13. Calculate EPI - Blue channel 405 nm laser power at objective lens.

    Repeat steps 5-12 of this guide using the Blue fluorescent plastic slide to calculate the EPI- Blue channel 405 nm laser power (p_{eb}) reaching the objective lens.

$$p_{eb} = \frac{int_{eb}}{(t_{eb} \times K)}$$

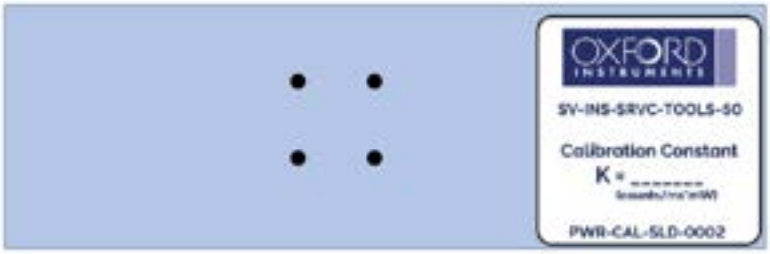


Figure 177: Blue fluorescent slide.

14. Calculate EPI - Yellow channel 561 nm laser power at objective lens.

Repeat steps 5-12 of this guide using the Yellow fluorescent plastic slide to calculate the EPI- Yellow channel 561 nm laser power (p_{ey}) reaching the objective lens.

$$p_{ey} = \frac{int_{ey}}{(t_{ey} \times K)}$$

Figure 178: Yellow fluorescent slide.

15. Calculate EPI - Red channel 638 nm laser power at objective lens.

    Repeat steps 5-12 of this guide using the Red fluorescent plastic slide to calculate the EPI- Red channel 638 nm laser power (p_{er}) reaching the objective lens.

$$p_{er} = \frac{int_{er}}{(t_{er} \times K)}$$

Figure 179: Red fluorescent slide.

Ensure all 4 calibration slides are put away into the XS-SMPL-TEST-SET box and store the box in a safe place where they can be easily found in the future.

In Windows, navigate to the directory that has been designated the Fusion Root Folder. Copy the raw .ims and metadata image files created during this functionality check and paste them into the C:\Program Files\Fusion\Support folder directory. Image files will be required for future comparisions of system performance.

Remember to reset the image file naming preferences back to your preferred settings after the functionality check has been completed.

Section 8 - Troubleshooting

Fault	Cause	Solution
The PC workstation will not power ON; the PC workstation power button is flashing white and orange.	The PC motherboard or some other hardware component of the computer has failed.	Contact your local Andor service representative
"USB device not recognized" error message appears in the Windows operating system of the PC workstation.	The USB cable connecting the BC43 to the PC workstation has become loose or unplugged.	Check and reconnect the USB cable on both ends to the instrument and the dedicated PCI card. Quit the software, power OFF the instrument, and restart the PC workstation. After the PC restarts, power ON the instrument, run the Fusion image acquisition software and check if the error message or connection issue has been resolved. If the problem persists, contact your local Andor service representative.
Fusion image acquisition software reports "Error LBLOC001: Communication lost with diagnostic Module".		
The front panel LED strip shows no light after powering on.		
On software startup, Fusion reports that a valid license cannot be found.	The Fusion license is locked to the PC. It is likely that the PC workstation name MAC address has been changed without following the recommended process (see Section "Changing the PC Workstation Name or Media Access Control (MAC) Address" on page 61	Contact your local Andor Sales or Technical Support team for assistance in resetting the Fusion license key.

Fault	Cause	Solution
The BC43 fails to connect to the Fusion image acquisition software.	The software was activated/run too soon after the instrument was powered on while the operating system was still setting up the camera driver.	Try quitting and restarting the Fusion acquisition software.
	The Fusion acquisition software icon was accidentally double clicked with the mouse from the Microsoft Windows operating system toolbar. This can cause two instances of the software to run potentially leading to hardware connection issues.	Open the Windows Task Manager and examine the Process Tab. Check if 2 instances of Fusion are running. If so, quit all instances of the software and make sure the program is properly re-activated/run.
	The previous instance of the Fusion acquisition software may have stalled upon shutdown and maintained a connection to the hardware, preventing the next restart of the software from making a proper connection to the instrument.	Quit the software and run the Windows Task Manager. Explicitly quit (End Task) any instances of the software that are still running under the Process Tab or the Details Tab, and then try restarting the software properly once more.

Fault	Cause	Solution
The Windows operating system environment becomes noticeably slow/ sluggish when Fusion runs a live preview or protocol acquisition.	A Windows 21H2 update (or an equivalent cumulative update) has caused one or more Windows Management Instrumentation (WMI) registry errors, which subsequently prevent Fusion from running properly after restart of the system.	Contact your local Andor service representative.
Fusion fails to load on startup and instead reports a Blake.Licensing related error message containing the phrase, "ManagementException – Not Found"		
The X-stage, Y-stage, or Z-stage fails to connect to software.	The Fusion acquisition software icon was accidentally double clicked with the mouse from the Microsoft Windows operating system toolbar. This can cause two instances of the software to run which can potentially lead to hardware connection issues.	Open the Windows Task Manager and examine the Process Tab. Check if 2 instances of Fusion are running. If so, quit all instances of the software and make sure the program is properly re-activated/run. If these steps don't work contact your Andor service representative.
	The previous instance of the Fusion acquisition software may have stalled upon shutdown and maintained a connection to the hardware, preventing the next restart of the software from making a proper connection to the instrument.	Quit the software and run the Windows Task Manager. Explicitly quit (End Task) any instances of the software that are still running under the Process Tab or the Details Tab, and then try restarting the software properly once more. If these steps don't work contact your Andor service representative.

Fault	Cause	Solution
Frequent Fusion acquisition software crashes.	The PC workstation monitor has been plugged into the wrong display port on the back panel of the PC workstation.	Check that the PC workstation monitor is connected to PC workstation via the HDMI to HDMI mini adapter cable and plugged into the devoted graphics processing unit (GPU) card (see "3.3.1 BC43 and PC Workstation Setup" on page 32). If not quit the software and power off the instrument. Then properly connect the monitor to the workstation. If these types of problems persist, contact your local Andor service representative.
The live preview or saved image appears blank/empty and every pixel in the image has a zero intensity value.	The connection between the system camera and main control board has failed.	Contact your local Andor service representative.
The confocal disk appears to partially block the widefield epifluorescence, brightfield, or differential phase contrast images.	- The system has accidentally been switched into quickboot mode. - The disk translation motor has failed.	Contact your local Andor service representative.
PC Workstation unexpected restarted during acquisition of a Fusion protocol	A Windows feature or security update has automatically downloaded and installed during the protocol, forcing a workstation restart	Check for Windows updates and immediately install BEFORE starting the experiment, or manually set an update installation delay time that exceeds the expected duration of the experiment. See Section "PC Workstation Operating System Notes and IT / Networking" on page 58.

Fault	Cause	Solution
The image focus changes significantly or the image field of view shifts significantly when a new objective lens is engaged.	A new objective lens has been added/ exchanged on the system without adjusting parfocality/parcentricity firmware settings	An additional procedure to adjust parfocality/parcentricity firmware settings is detailed in "Install an Objective Lens" on page 117
The 'Find Coverslip' button is greyed out in the Fusion interface for a dry objective (other than 2x).	An objective lens not listed in Fusion's configuration files is being used or an unsupported objective lens has been loaded into the system.	Contact your local Andor service representative to arrange a software update or a configuration file update
The 'Add Oil' dialogue sequence did not appear when activating an oil immersion objective lens in Fusion.		
The XY stage movement lags behind the movement of the joystick or the acquisition software seems to run slow.	The instrument has been plugged into the wrong USB port on the back panel of the PC Workstation.	Check that the BC43 instrument is connected to back panel of the PC workstation via the USB 3 cable into the devoted USB 3 port of the PCI express card marked with an orange dot (see "BC43 and PC Workstation Setup" on page 54). If not quit the software and power off the instrument. Then properly connect the instrument to the workstation.
The XY-stage and/or Z-focus drive lags responses to 3D joystick inputs.	The Windows operating system may fail to recognize the 3D Connexion Spacemouse joystick the first time it is plugged into a USB port on the PC workstation that it has not been plugged into before.	Quit the Fusion acquisition software and power OFF the instrument. Restart the PC workstation. After restart, power ON the instrument and run the Fusion acquisition software again. The XY-stage and Z-focus drive should now respond properly to joystick inputs.

Fault	Cause	Solution
The XY stage overshoots the input motions of the 3D Connexion Spacemouse joystick.	The joystick has been plugged into a USB 2.0 port instead of a USB 3.0 port on the PC workstation.	Quit the Fusion image acquisition software, Power OFF the BC43. Plug the joystick into a USB 3.0 port on the PC workstation and then restart the PC workstation. Once restarted, power ON the BC43 and run the Fusion image acquisition software.
Acquired images exhibit an artefact that manifests as 'rain drops' or dispersed spots that superimpose the expected image.	The detector vacuum seal has failed and moisture has leaked into the detector sensor chamber.	The camera must be replaced. Contact your local Andor service representative.
The left/right joystick buttons on the 3DConnexion Spacemouse are unresponsive in Fusion	The joystick buttons are not assigned to operate with the image acquisition software after the creation of a new Windows login account on the PC station.	Configure the joystick buttons on the new Windows login account (see"Connect and Configure the 3D Connexion Spacemouse" on page 55).

Fault	Cause	Solution
No laser light observed exiting from the microscope objective lens or no transmitted white LED light projected onto the sample; sample not illuminated.	The live preview mode is not active, or no protocols are running.	Activate Live Preview mode or run a Protocol in the Fusion acquisition software interface.
	The system interlock has been tripped.	Close the top lid cover and/or front access panel door. After the top lid has been open for 5 minutes with the live preview mode running, all laser light and transmitted light will be shuttered. Closing the lid completes the interlock circuit and turns on the laser light or transmitted light again at unattenuated strength. If you desire to view the specimen again, the lid can be lifted and viewed in safe mode with attenuated light conditions. After 5 minutes, the interlock will trip again.
	Ensure laser intensity set above 0%.	Activate Live Preview mode or run a Protocol in the Fusion acquisition software interface. The channel laser must be explicitly turned on by the user in the Fusion acquisition software interface, and the laser intensity slider must be set to any value above 0% to turn on when live preview mode is activated.
	Laser engine thermal warning.	Contact your local Andor Sales or Technical Support team to order a replacement fan and arrange service call to assist with this error.
	One or more lasers in the laser engine has failed.	Contact your local Andor Sales or Technical Support team to arrange a laser engine replacement.
	The transmitted light turret LED has failed.	Contact your local Andor Sales or Technical Support team to order a replacement turret and optionally arrange service call to assist with this procedure.

Fault	Cause	Solution
Sample is illuminated with laser light, but little to no fluorescence emission light is detected on the camera, image signal is weak and noisy or blank/absent.	The top lid cover is open, full laser strength has been attenuated.	Close the top lid cover. Closing lid will stop the safe mode laser attenuation. Re-adjust laser power and exposure time as needed.
	The expression level of the fluorescent probe/-contrast agent/stain is too low or did not label the structure of interest within the sample properly. The sample may be bleached already from previous imaging with all fluorophores depleted.	Consult the fluorescent probe's manufacturer's protocol for sample labelling troubleshooting tips or contact the manufacturer directly for additional advice regarding optimal staining. Use a positive control sample with the same fluorescent probe and labelling protocol to verify and validate the experiment.
	An empty FOV of the sample is being observed.	Laterally scan the sample with the microscope stage to find a part of the sample that is fluorescent or use the microscope's transmitted light imaging modalities (brightfield or DPC) to locate the specimen of interest.
	The sample is not in focus.	Click the "Find Coverslip" option in the Fusion software interface (not available for 2x).
	The wrong imaging channel is being used to view the sample.	Ensure that the correct imaging channel is being used to observe the fluorescent contrast agent / probe of interest. Consult the fluorescent probe's manufacturer's specification sheet to verify that the imaging channel matches the excitation/emission spectrum of the probe.
	The camera exposure time is too low.	Holding the excitation laser power fixed, try increasing the camera exposure time 2-fold, 5-fold, or 10-fold until the image signal strength approaches an acceptable level below the saturation threshold of the camera pixels.

Fault	Cause	Solution
	The laser power is too low.	Holding the camera exposure time fixed, try increasing the excitation laser power until the image signal strength approaches an acceptable level below the saturation threshold of the camera pixels. If the sample begins to rapidly photobleach or the signal level does not significantly improve by the time maximum laser power is reached, try increasing the camera exposure time too.
	Image intensity mapping may be incorrect or out of range.	Enable 'Auto Mapping' in the Fusion acquisition software interface to assist finding signal.
	The imaging optics may be contaminated and require cleaning.	Clean the objective lenses. Check that the FSL module dichroic mirror and clean if necessary, following the procedure "5.1.5 Cleaning the Focus Seek and Lock Module" on page 69. Check the front chassis infinity space sealing window and clean if necessary, following the procedure "5.1.3 Cleaning Objective Lenses" on page 67.
	The laser light coupling into microscope optics is too low.	Contact your local Andor Sales or Technical Support team to arrange a laser engine replacement.
	The camera fan has failed.	Contact your local Andor Sales or Technical Support team to order a replacement camera fan and arrange a service call to assist with this procedure.
	Fluorescent probe in sample does not optimally match the excitation wavelength and/or emission filter spectrum.	Please refer to "Technical Specifications" on page 215 on page 92 for more information.

Fault	Cause	Solution
Sample is illuminated with laser light, camera pixel intensity is saturated.	Laser illumination power or camera exposure time is set too high.	Reduce the laser power and or the camera exposure time in the Fusion image acquisition software interface until the image signal strength is uniformly below the pixel intensity saturation threshold limit.
	Sample is exposed to room light.	Close the top lid cover.
	The camera fan has failed.	Contact your local Andor Sales or Technical Support team to order a replacement camera fan and arrange a service call to assist with this procedure.
The image has a "speckled" appearance that slowly changes with time.	Unit homogeniser is not functioning correctly.	Contact your local Andor Sales or Technical Support team to arrange a laser engine replacement.
The sample will not come into focus or the objective lens is crashing into the sample coverslip.	The objective lens is contaminated and requires cleaning.	Clean the objective lens.
	The sample holder is not seated properly.	Follow the procedure to correctly insert the sample holder "Inserting/Removing the Sample Holder" on page 101.
	The correct sample holder type has not been selected in software.	Ensure the "Incubator" option is only active when using the Okolabs stage-top incubator.

Fault	Cause	Solution
The Find Coverslip option can't find the sample focus; The sample focus is drifting even when the Focus Lock option is active in a protocol.	The sample substrate is not compatible with the objective lens.	Plastic bottom substrates are permissible for dry (non-immersion) objective lenses only. Change the objective lens to a dry objective or use glass-based #1.5 (0.16 - 0.19 mm) coverslips to mount the sample. Use recommended sample substrates (See table in "Recommended Sample Substrates" on page 46).
	The FSL module may require cleaning	Follow the procedure to clean the FSL module dichroic mirror (refer to section).
	The front chassis infinity space sealing window may be contaminated and require cleaning.	Follow the procedure to clean the front chassis infinity space sealing window "Cleaning the Objective Lenses" on page 130.
	A new or replacement FSL module has been loaded into the instrument without proper calibration in the instrument control software.	Contact your local Andor Technical Support team to arrange for proper FSL calibration.
	The FSL module is not functioning correctly.	Contact your local Andor Sales or Technical Support team to arrange a FSL module replacement.
The stage not responding correctly to XY joystick commands, or the z-stage is too sensitive/not sensitive enough to focus changes.	The wrong objective lens has been inserted into the objective nosepiece position.	Follow the prescribed procedure for removing/inserting objective lenses from the unit "Install an Objective Lens" on page 117.
	Navigation speed settings not optimally set.	In Fusion go to Navigation Preferences and adjust the speeds and nudge sizes. For optimal navigation preference settings please see the BC43 Fusion guide.

Fault	Cause	Solution
The front access panel door falls uncontrollably when opened.	The front access panel door drawbridge mechanism has failed.	Replace the front access panel door drawbridge mechanism using the prescribed procedure "Replacing Front Access Panel Door" on page 157. Contact your local Andor Sales or Technical Support team to order a replacement drawbridge mechanism and optionally arrange a service call to assist with this procedure.
The top lid cover won't stay open and/or falls too fast when closing.	The top lid cover gas strut has failed.	Contact your local Andor Sales or Technical Support team to arrange top lid cover gas strut replacement.
Some sample features near tile borders appear twice / doubled in the stitched image of a montage protocol.	The montage Overlap parameter has been set too low.	Try increasing the montage overlap parameter by 5-10% and repeat the image acquisition protocol.
Fusion image channels disappear, or unknown image channels appear in channels list.	The Fusion channels configuration file has become corrupted.	Contact your local Andor service representative to assist and restore the default channels.
The confocal image does not come into focus evenly; the confocal image intensity sweeps across field of view in a Z-scan. The confocal image intensity discontinuously changes at the tile borders of a confocal montage	The specimen slide is not level on the XYstage or the coverslip rests at an angle with respect to the XY-stage.	Level the XY-stage insert according to the instructions in "Adjusting the Sample Holder for Sample Tilt" on page 103

Fault	Cause	Solution
The PC workstation monitor is exhibiting screen flicker	The mini DisplayPort to DisplayPort adapter has failed and needs to be replaced.	Contact your local Andor service representative.
Part of the PC workstation monitor screen unresponsive to mouse clicks.	Windows operating system bug.	

Appendix

The following sections contain information on product specifications, including technical, environmental, mechanical and electrical specifications. In addition, detailed mechanical drawings are presented.

Appendix A: Technical Specifications

Microscope Unit	BC43
Imaging Modes	**High-speed confocal** Full frame = 15 fps; Binning 2x2=20 fps, Binning 4x4=20 fps Cropped 2048x1024 =19 fps; Cropped 1024x1024 =30 fps, 512x512=44 fps **Widefield epifluorescence** Maximum Frame Rate = 15 fps **Transmitted light brightfield and Differential Phase Contrast** BF Maximum Frame Rate = 15 fps Fast DPC Maximum Frame Rate = 3.7 fps HQ DPC Maximum Frame Rate = 1.8 fps
Imaging Methods	Single colour, multicolour, z-stacking (volume), time-lapse, Optional modules: multi-field/multi-well, area scan (montage) with tile stitching.
ClearView™ GPU	Clears image of non-specific sample back-ground signal and improves resolution beyond the normal optical limits.
Camera	
Resolution	6.5 µm pixel; 2040x1992 pixels (4.1 MP)
QE	Up to 82%
Field of view (mm)	18.4 mm (diagonal)
Cooling	0°C
Images	16-bit, monochrome
Illumination	
Fluorescence	Fixed configuration of 405 nm, 488 nm, 561 nm, 638 nm
Safety Class	Class I under normal operation; Class II for access & maintenance.
Transmitted light	Broad spectrum visible light LED
Emission Filters Centre wavelength/Bandwidth (nm)	Blue 445/20, Green 529/24, Orange 595/31, Red 708/75
Optics (Objectives)	
Objective Lens Nosepiece	Motorised 5 position turret

Optics (Objectives)	
Objective Magnifications	Supplied with 2x objective for sample overview. Select additional supported objective lenses from 10x to 100x magnification.
Precision motorised x,y stage	Travel Range = 110 mm x 80 mm Resolution = 100 nm
Z-Control & Focus	Range = 14.5 mm Resolution = 100 nm Repeatability = +/-10 nm
Autofocus "Seek & Lock" Technology	Finds focal plane for new sample, maintains focus stability during time-lapse experiments. Time to Find coverslip "Seek" =~15 secs Time to check focus in time series "Lock" = ~8 secs See "Accessories" on page 43for list of supported objectives.
Sample Vessels Supported	Glass slides (25 by 75 mm); culture dish (35 mm diameter); Multiwell plates (6, 12, 24 & 96); Multiwell chamber coverslip (2, 4, 8). Recommended Manufacturers (Mattek, Greiner) Supported Manufacturers (Ibidi, Labtek), see "Recommended Sample Substrates" on page 46.
Incubation (option)	Stage-top incubator. Sliding lid for easy sample access and exchange. Objective heater for oil-immersion objectives.
Workstation	
PC	Windows 11™ operating system Fusion control and Imaris Quant software 64 GB (2x32GB) DDR5 RAM 512 GB SSD Boot drive 16 GB Graphics Card 2 TB Image data storage (option to add more)
Monitor	Monitor 27 inch QHD (2560 x 1440)

Mechanical Specifications

BC43	
Weight (BC43 unit only)	64 kg
Weight (External Power Supply)	0.85 kg
Dimensions	See Mechanical Drawings

Environmental Specifications

Location to be used	Indoor use only
Operating temperature	18-25°C ambient
Storage temperature	0°C to 50°C
Operating relative humidity	<70% (non-condensing)
Pollution degree	Pollution degree 2. Normally only non-conductive pollution occurs. Occasionally, however, a temporary conductivity caused by condensation must be expected.
Cooling vent clearance	Do not cover during operation. Allow 100 mm clearance at air vents. Allow 250 mm clearance at air vents.

Appendix B: BC43 Laser Power Specifications

Wavelength / nm	Laser output power at objective lens back aperture (100%)				
	Lid open*		Lid closed		
	Confocal illumination mode		Confocal illumination mode		
	typ / mW	max / mW	min / mW	typ / mW	max / mW
405	NA	NA	6	10	13.5
488	0.1	0.2	4	6.5	8.5
561	0.2	0.3	4.5	6.5	9
638	0.2	0.3	6	8.5	11
*Note 405 nm laser off when lid is open.					

Wavelength / nm	Laser output power at objective lens back aperture (100%)				
	Lid open*		Lid closed		
	Widefield/ Epifluorescence illumination mode		Widefield/ Epifluorescence illumination mode		
	typ / mW	max / mW	min / mW	typ / mW	max / mW
405	NA	NA	12.5	20	27
488	0.2	0.3	8.5	13	16.5
561	0.4	0.6	9.5	13	17.5
638	0.4	0.6	12	17	21.5
*Note 405 nm laser off when lid is open.					

Other Power Values	
Worst-case accessible emission at objective*† / mW	0.6
Incorporated laser module worst-case emission at fibre output‡ (internal to unit and inaccessible) / mW	Total = 94, Condition 3* = 6.7

Beam Divergence	
Beam divergence range / mrad	Widefield/Epifluorescence: 4.4 to 220 Confocal: 90 to 2300
Beam divergence at objective turret with no objective fitted / mrad	Widefield /Epifluorescence: 50 Confocal: 260
Incorporated laser module beam divergence at fibre output (internal to unit and inaccessible) / degrees	13.8

Beam Diameter	
Beam diameter at objective aperture/ mm	18.4 divided by magnification
Beam diameter at objective turret with no objective fitted / mm	0.62 (Widefield/Epifluorescence), 8.2 (Confocal)
Incorporated laser module beam diameter at fibre output (internal to unit and inaccessible) /	2.5

Beam Diameter	
mm	

* IEC 60825-1 Condition 3 measurement which is using a 7 mm limiting aperture at 100 mm from laser aperture to simulate the eye pupil at its widest extent at the standardised minimum reasonable distance to approach the hazard.

† This is for Epifluorescence Illumination Mode using a 20x objective and the worst-case wavelength of 561 nm. This compares with the Class 2 AEL of 4.3 mW for that objective and our laser radiation field characteristics. It is very hard to intercept the laser beam with the eye without using a reflective surface to redirect it.

‡ This is for the worst-case wavelength of 405 nm.

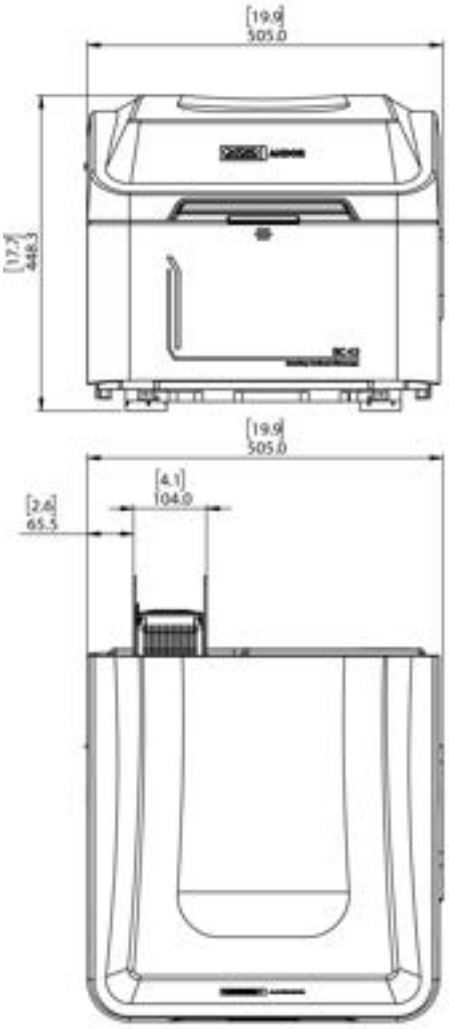
Appendix C: Electrical Power Specifications

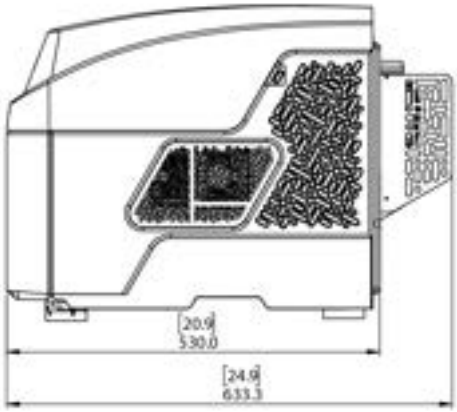
	BC43
Mains Input for Supplied External Power Supply	100 – 240 VAC, 50 – 60 Hz
Power Consumption (inc. External Power Supply)	12 W (standby) 60 W (typical)•[1] 95 W (max)
Voltage Rating	24 V
Current Rating	6 A
Mains Overvoltage Category	CAT II An overvoltage category of CAT II means that the equipment is designed to cope with transient voltages above the rated supply that would be experienced by any product connected to a standard single-phase mains socket in a building.

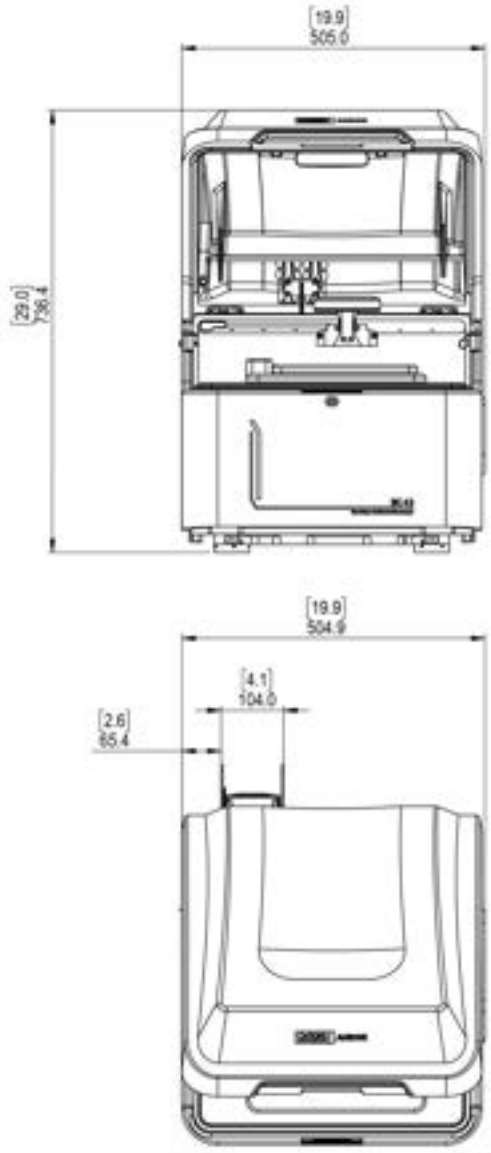
External AC/DC Power Supply Requirements

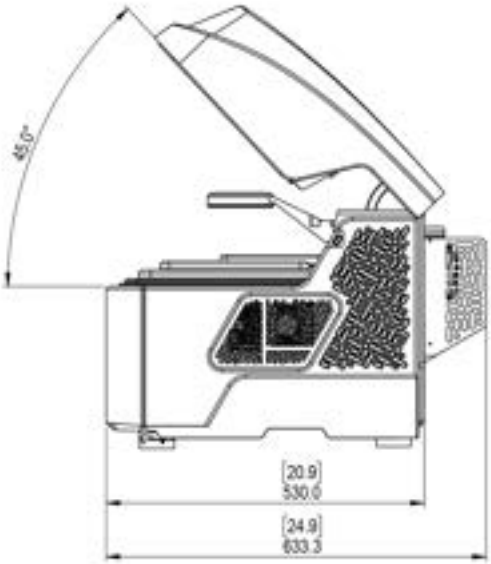
	BC43
Low Voltage Supply Input	24 V
Low Voltage Supply Current	See current rating above
Low Voltage Supply Cable Plug	KYCON KPPX-4P
Low Voltage Supply Cable Plug Insertion View	
Low Voltage Supply Pin Connections	Pins 1 & 2 +24 V Pins 3 & 4 GND
Low Voltage Supply Product Socket	Kycon KPJX-4S
Low Voltage Supply Product Socket Insertion view	
Ripple	480 mV Max.
Safety	Certified to IEC 62368-1 in accordance with local safety regulations and meets the reinforced insulation from mains requirement of IEC 61010-1
Environmental	Ensure that the EPS meets the environmental specification of the overall product

Appendix D: Mechanical Drawings






19.9
505.0
29.0
736.4
19.9
504.9
4.1
104.0
2.6
65.4


45.0°
20.9
530.0
24.9
633.3

Appendix E: Connection Diagrams for Previous PC Models

Connection diagram for earlier PC models shipped with BC43.

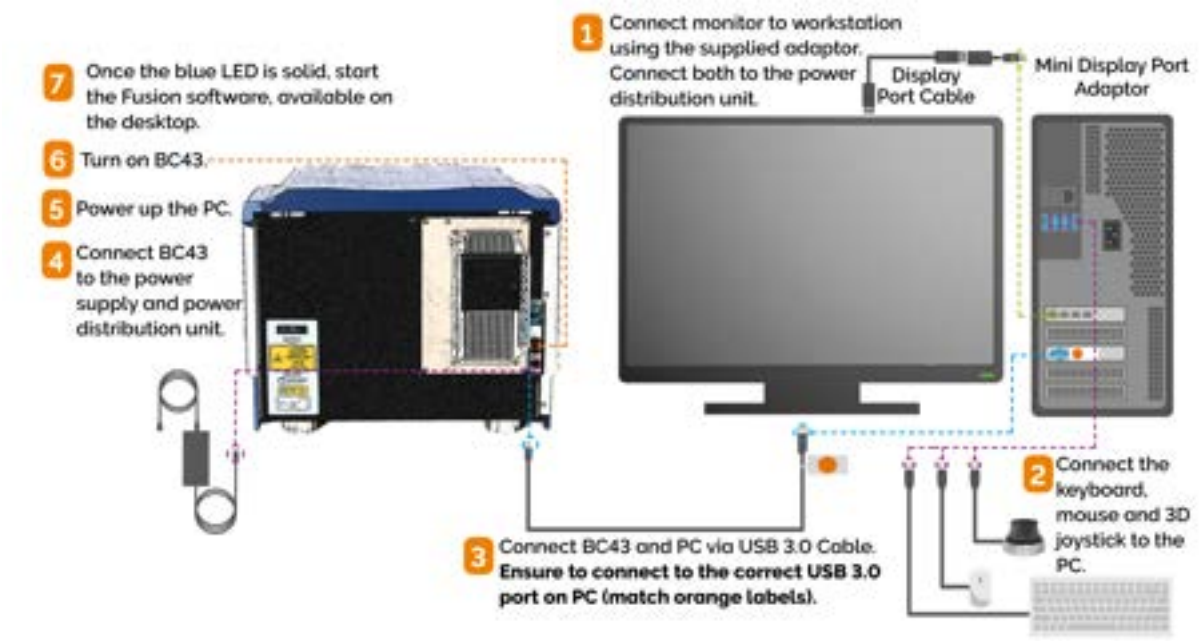


Figure 180: Connection diagram between PC, BC43 and accessories for PC models Dell 3640 and Dell 3650.

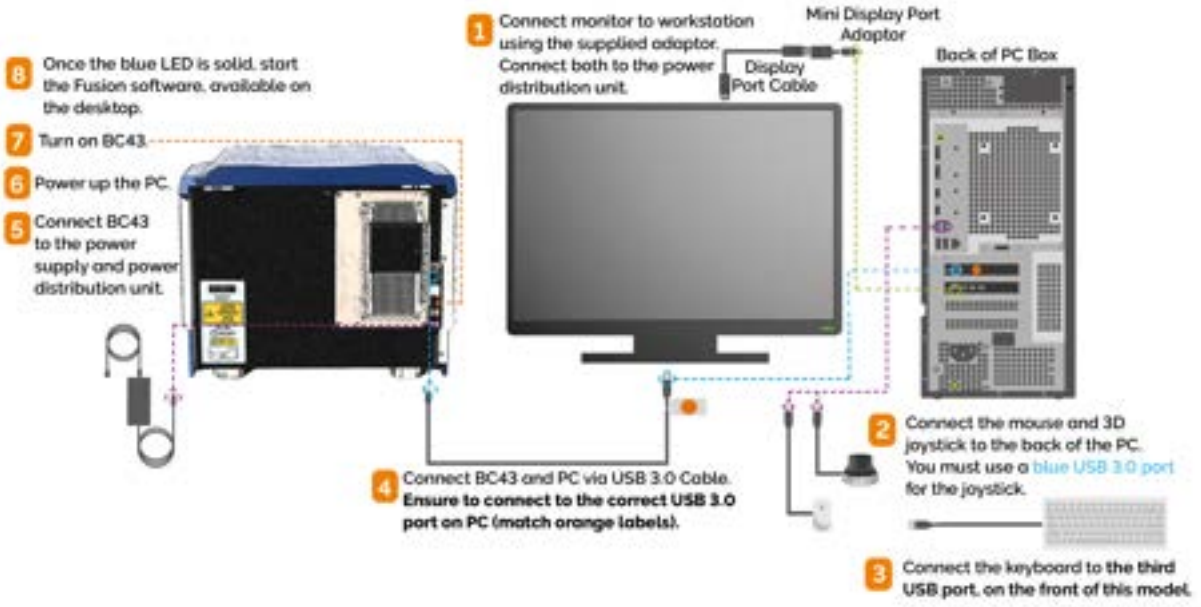


Figure 181: Connection diagram between PC, BC43 and accessories for PC model Dell 3660.

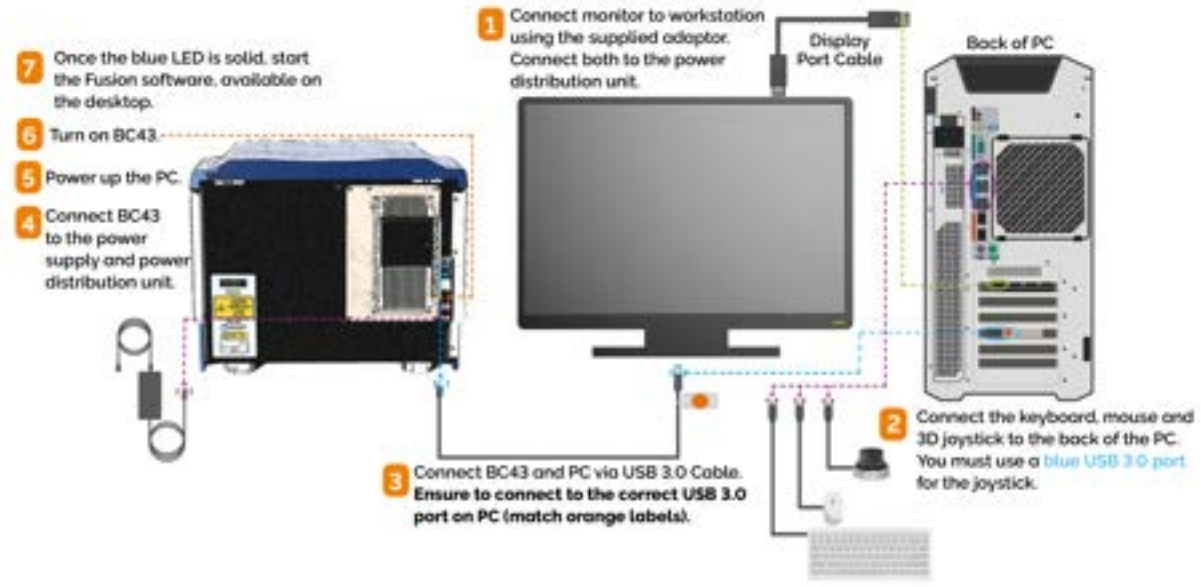


Figure 182: Connection diagram between PC, BC43 and accessories for PC model HPZ8G4.

Appendix F: Other Information

Terms and Conditions of Sale and Warranty Information

The terms and conditions of sale, including warranty conditions, will have been made available during the ordering process. The current version for the US is available here, for all other regions (except Japan) please click here.

EU/UK REACH Regulation Statement

Andor's EU/UK REACH Regulation statement is available at the following link.

Waste Electronic and Electrical Equipment

The company's statement on the disposal of WEEE can be found in the Terms and Conditions.

Appendix G: China RoHS Hazardous Substances Declaration

Hazardous Substance: 有害物质						
Component Name 部件名称	Lead (Pb) 铅	Mercury (Hg)汞	Cadmium (Cd)镉	Chromium VI Compounds (Cr6+)	Polybrominated Biphenyls (PBB)	Diphenyl Ethers (PBDE)
Printed Circuit Board Assemblies (Surface-mount Resistors and Capacitors, and Brass Connectors) 路板组件 (表面贴装电阻器和电容器，以及黄铜连接器)	X	O	O	O	O	O
Hex Stand-offs (see image in table below) 六角隔撑	X	O	O	O	O	O
Screw Locks (see image in table below) 螺丝锁定	X	O	O	O	O	O
Kinematic mount 架 六角隔撑	X	O	O	O	O	O
Metal Rail 六角隔撑	X	O	O	O	O	O
Metal Rail Reader 导轨引头	X	O	O	O	O	O
Computer Hard Drive 电脑硬盘	X	O	O	O	O	O
Computer USB3 Card 口卡	X	O	O	O	O	O
Optical Fibres 光纤	X	O	O	O	O	O
Camera DC power socket 相机直流电源插座	X	O	O	O	O	O

Hazardous Substance: 有害物质						
Component Name 部件名称	Lead (Pb) 铅	Mercury (Hg)汞	Cadmium (Cd)镉	Chromium VI Compounds (Cr6+)	Polybrominated Biphenyls (PBB)	Diphenyl Ethers (PBDE)
Camera DC power supply connector 相机直流供电接口	X	O	O	O	O	O
All other parts 其余配件	O	O	O	O	O	O

This table was developed according to the provisions of SJ/T 11364

本表格依据SJ/T 11364 的规定编制

O - The content of such a hazardous substance in all homogeneous materials of such a component is below the limit required by GB/T 26572

表示该有害物质在该部件所有均质材料中的含量均在O -表示该有害物质在该部件所有均质材料中的含量均在GB/T 26572 规定的限量要求以下

X - The content of such a hazardous substance in a certain homogeneous material of such a component is above the limit required by GB/T 26572

X - 表示该有害物质至少在该部件的某一均质材料中的含量超出GB/T 26572 规定的限量要求

Component Name 部件名称	
Hex Stand-offs 六角隔撑	
Screw Locks 螺丝锁定	

Component Name 部件名称	
Kinematic mount 架 六角隔撑	
Metal Rail 六角隔撑	
Metal Rail Reader 导轨引头	
Camera DC power socket 相机直流电源插座	
Camera DC power supply connector 相机直流供电接口