2. Click the [Calibrate] button.
   » The magnification test scan for the monochrome camera is identical with the magnification test scan for the color camera. See Magnification Test Scan on page 183.

   The resulting value should be within a deviation of +/- 0.1 degree.

If the calibration value is not within the standard go back to the [Camera Alignment] calibration and readjust the rotation. This will also effect the shading correction, however. Refer to chapter Shading Correction (Bright-field) on page 162 to redo the shading correction.

24.7 Shading correction for fluorescence observation methods

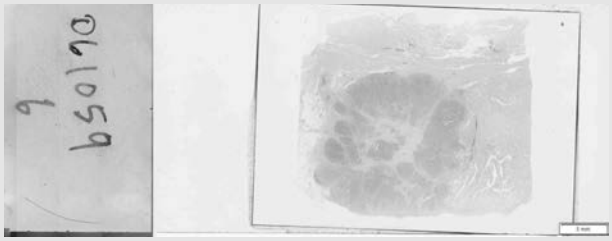
The shading correction for DAPI, FITC, CY3 and Cy5 can be done on a 'normal' thin, homogenous and large H&E sample using the auto-fluorescence as shown in the example below.

The slide for the shading correction is not part of the system. However, any H&E sample with a minimum size of 1.5 cm x 1.5 cm can be used.

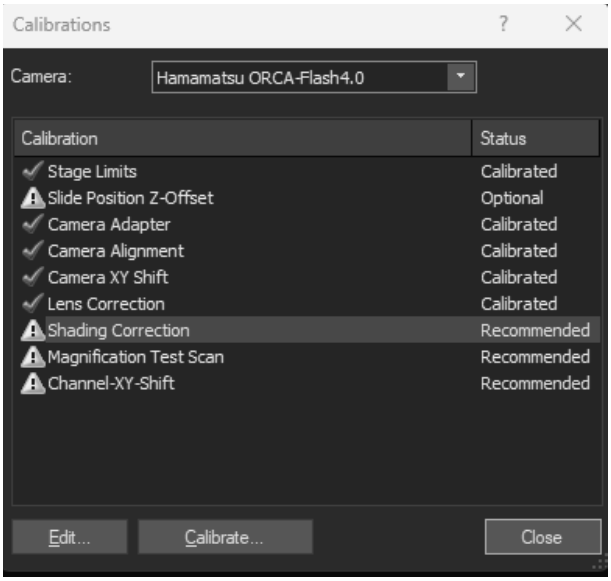
To perform the shading correction for CY7 a real CY7 stained sample is necessary as CY7 does not show any autofluorescence.

However shading correction is also sample dependent. Especially the thickness of the sample might have an influence on the correction.

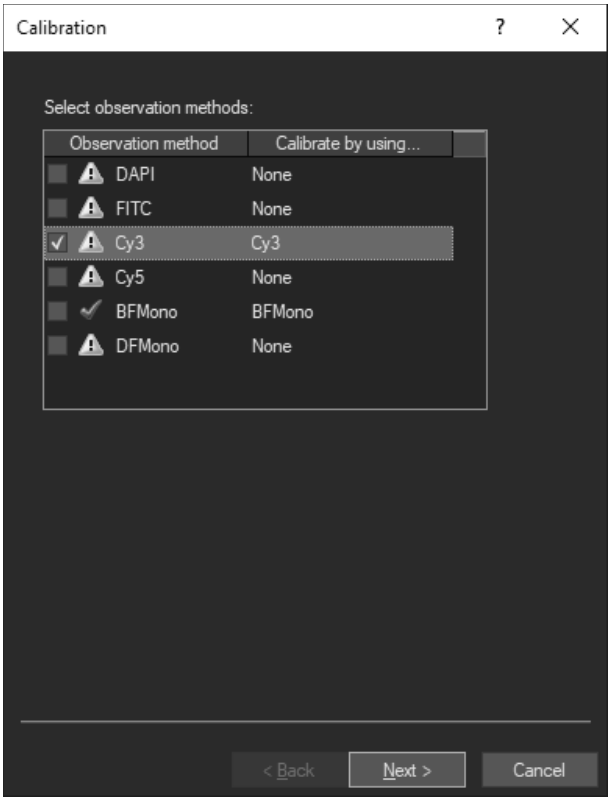
It could be necessary to repeat the shading correction if different samples are used.



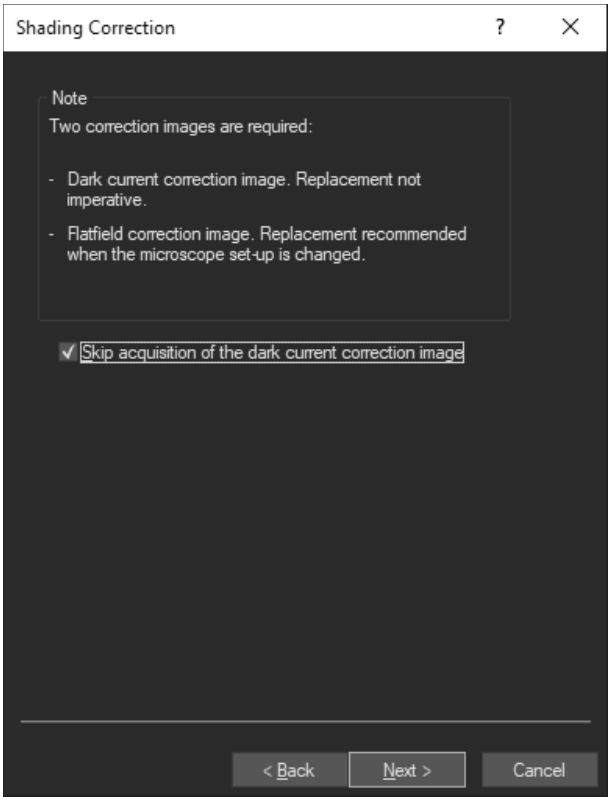
1. Put a similar slide into a tray and load the tray.
2. In the [Calibrations] dialog box select [Shading Correction] entry.



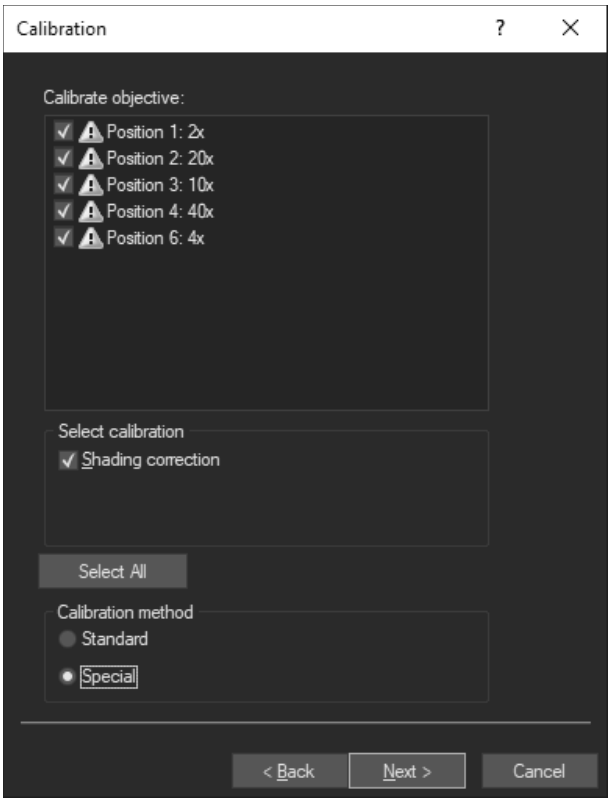
3. Select e.g. the [Cy3] observation method and click [Next].



4. Always skip the acquisition of the dark current correction image.

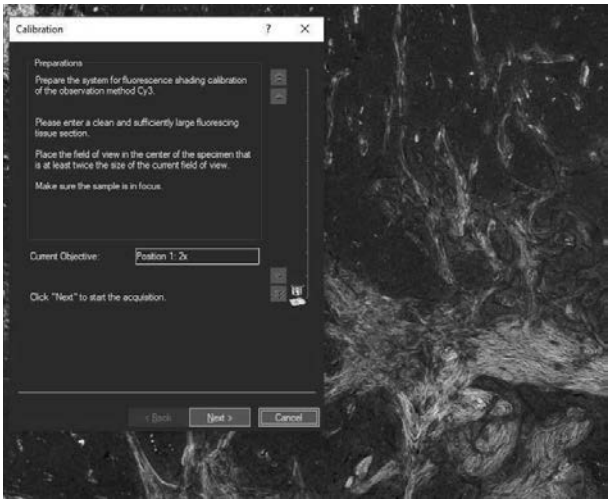


5. Select all objectives. Immersion objectives are calibrated in the same wizard. However the calibration wizard will calibrate all dry objectives first and subsequently select the immersion medium objective(s).
6. It is very important to select the calibration method [Special] in the [Calibration method] group. Only the calibration method [Special] will work on a real sample. The calibration method [Standard] requires different samples and is less effective. Also make sure that the [Shading correction] check box is selected.

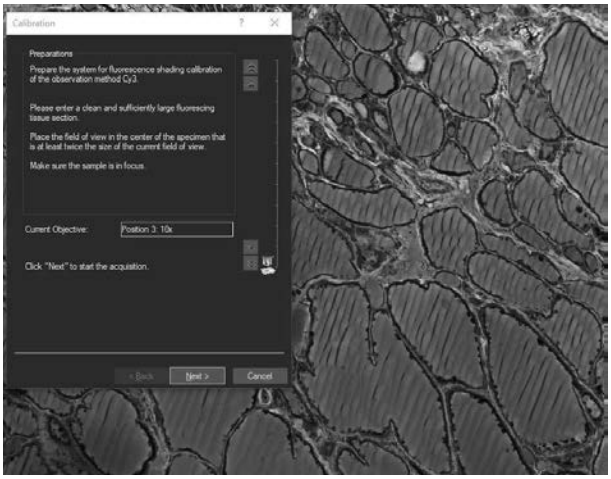


7. Proceed with [Next].
   » The system switches into live mode.

8. Select an area on the slide large enough for a 2x field of view and focus the sample via 'Ctrl' + mouse wheel and proceed with [Next].



For objectives equal or higher than 10x try to find a sample area which is dense and homogenous.



9. Repeat these steps for all other selected objectives.
10. Continue the shading correction for all other fluorescence observation methods.

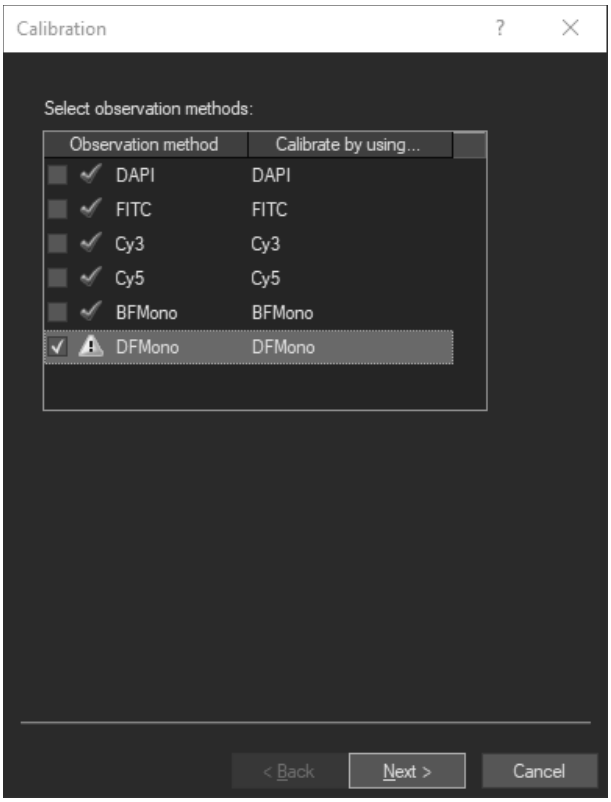
ATTENTION

In case you have calibrated an immersion medium objective as well clean the objective and remove immersion medium residues from the calibration slide.
See Cleaning the immersion objective on page 226.

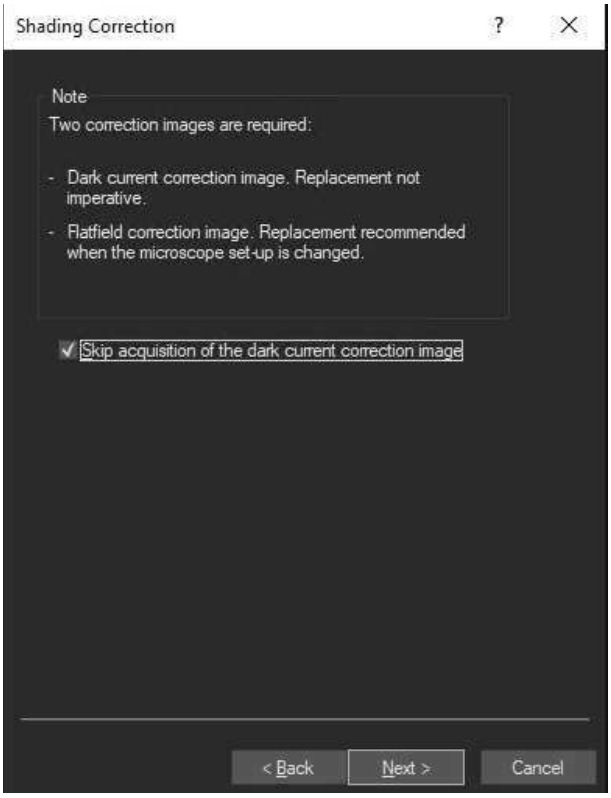
24.8 Shading correction for darkfield (DFMono)

- The darkfield observation method can be used with objectives < NA 0.8 only.
- For the default configuration of the VS200 system it can be done for the 4x and 10x objective only.
- The shading correction can be done on a thin homogenous sample with a minimum size of 1.5cm x 1.5cm (same as shading correction for fluorescence images).

1. In the [Calibrations] dialog box, select the [Shading Correction] entry and click the [Calibrate] button.
2. Select the [DFMono] observation method and proceed with [Next].



3. Skip the acquisition of the dark current correction image. Proceed with [Next].

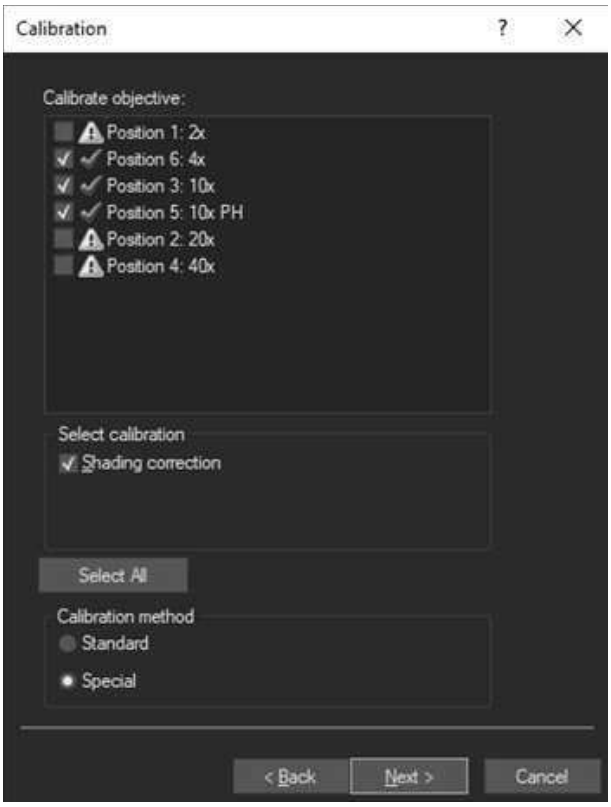


4. Select the check box for the 4x and 10x objective.

   Make sure that the [Shading correction] check box is selected.

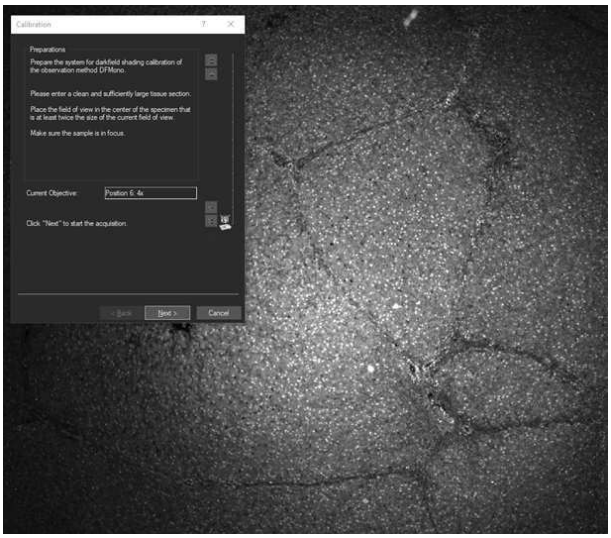
   Select the [Calibration method] > [Special] option.

   Proceed with [Next].

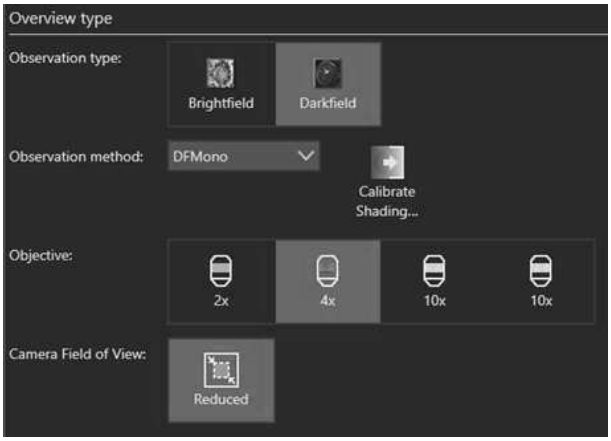


» The system switches into live mode.

5. Select an area on the slide which is homogenous on the current and adjacent field of view in all directions. Focus the sample. Use [Ctrl] and the mouse wheel to focus.

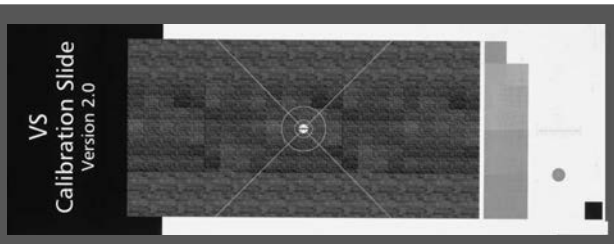


6. Proceed with [Next].
7. Repeat these steps for 10x.
8. Click the [Finish] button to finalize the shading correction.
9. During the scan process it is important to select the [Camera Field of View] > [Reduced] button when using the 4x objective to avoid vignetting.



10. If different samples are used it might be necessary to perform the shading correction again using the [Calibrate Shading Correction] button during the scan process.

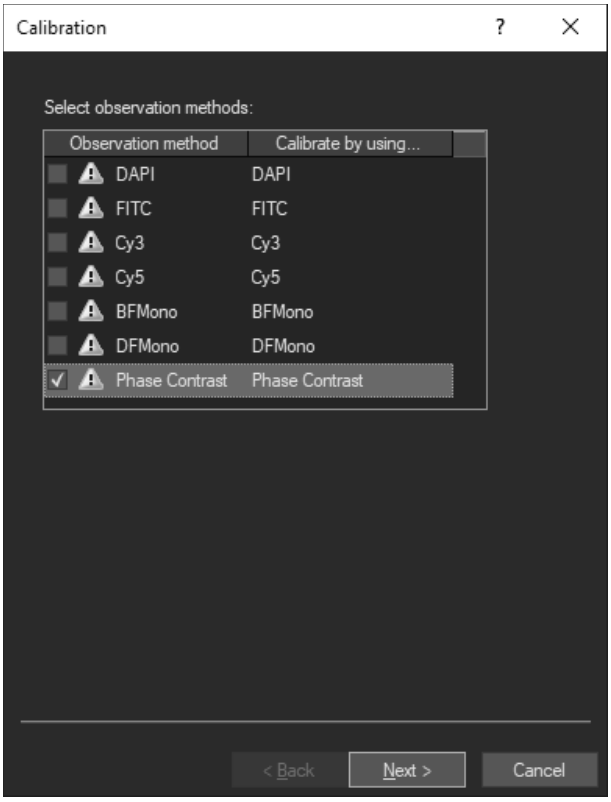
24.9 Shading correction for Phase Contrast (PH)



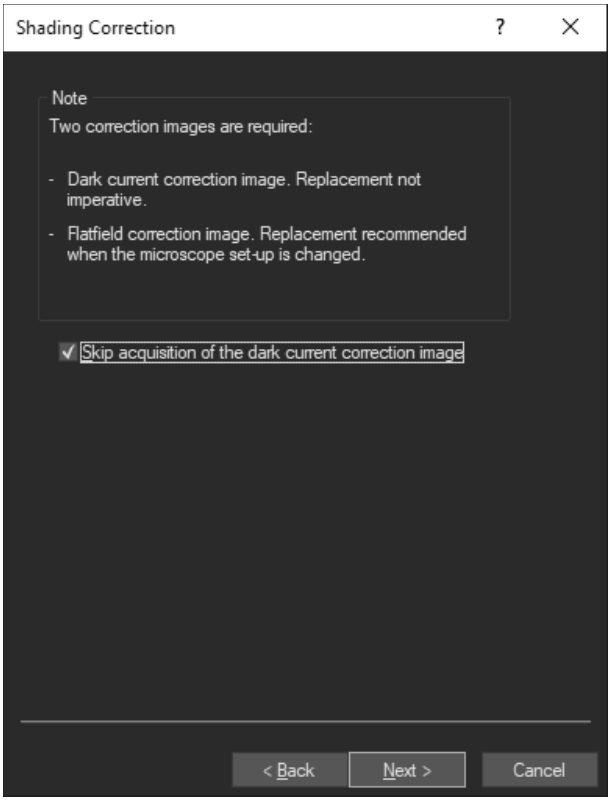
To perform the shading correction, use the calibration slide delivered with the system.

1. In the [Calibrations] dialog box select the [Shading correction] entry.
2. Click the [Calibrate] button.

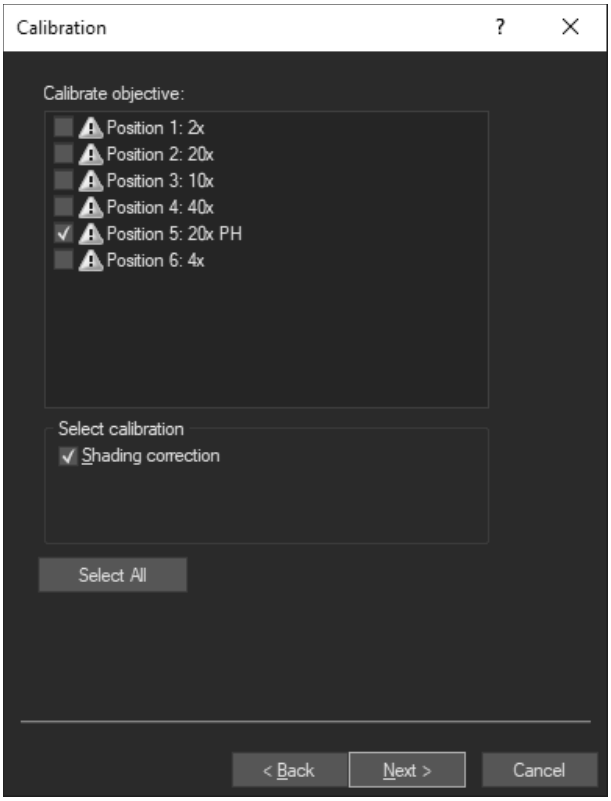
3. Select the [Phase Contrast] observation method.



4. Skip the acquisition of the dark current correction image.

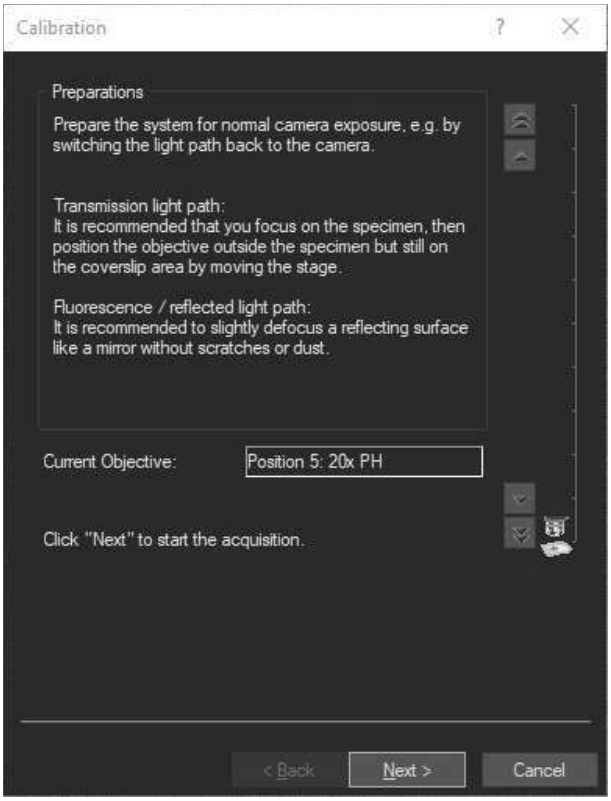


5. Only select the phase contrast objective(s). Make sure that the [Shading correction] check box is selected.

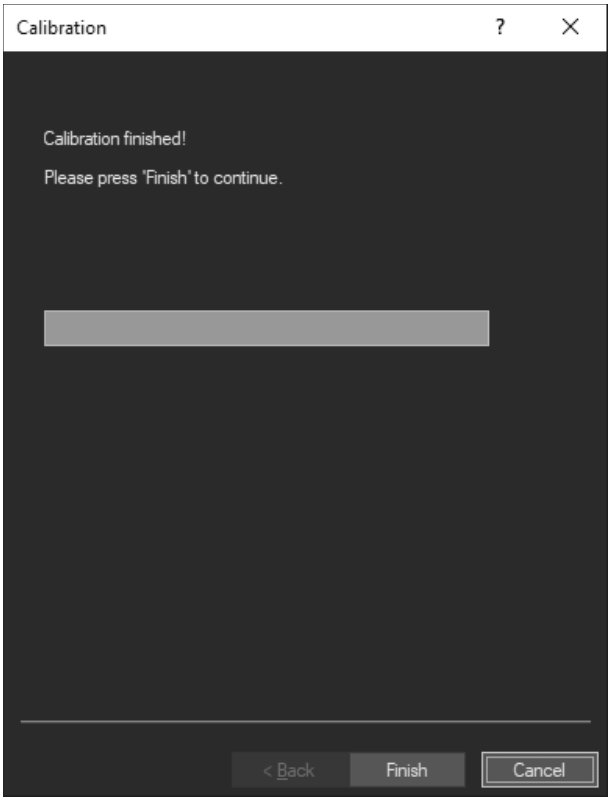


6. Focus on the calibration slide structure. Then go to the right side of the calibration slide to search for a very clean area to acquire the shading image.

7. Proceed with [Next].



8. Finalize the process by clicking the [Finish] button.



24.10 Channel-XY-Shift

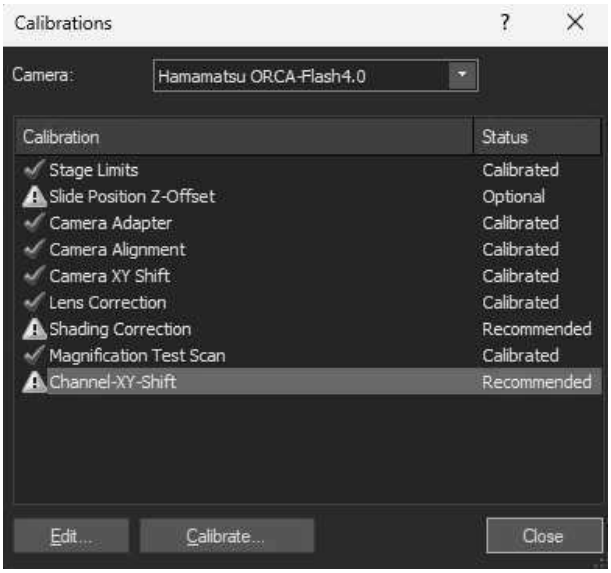
The channel-XY-shift calibration is only needed if multiple fluorescence filters or dichroic mirrors in the IX3-RFACA are used.

If the customer's system setup only contain one dichroic mirror in the IX3-RFACA, this calibration has not to be performed.

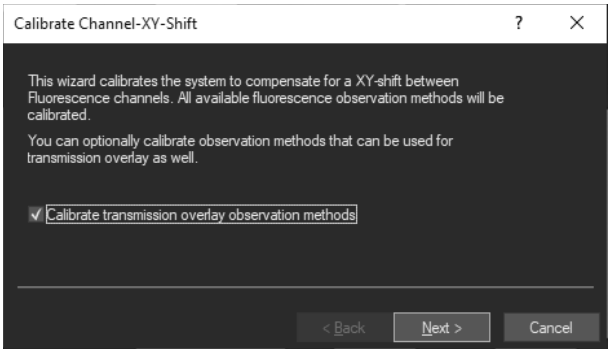
The slide for the XY shift correction is not part of the system. However, any H&E sample with a minimum size of 1.5 cm x 1.5 cm can be used.

It is important to correct a XY pixel shift induced by the different filter cubes.

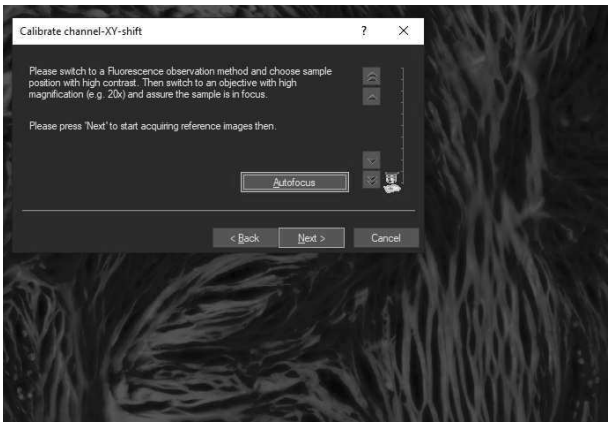
1. In the [Calibrations] dialog box select the [Channel-XY-Shift] entry.
2. Click the [Calibrate] button.



3. Select the [Calibrate transmission overlay observation methods] (bright field) check box.

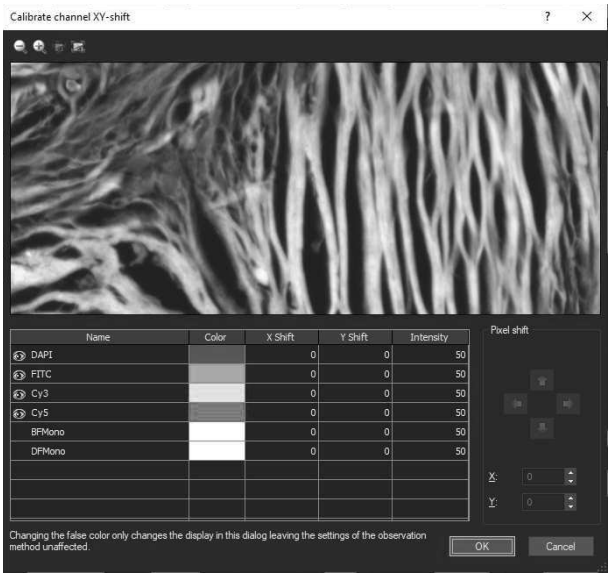


4. Put in a customer fluorescent sample which contains dyes for all of the fluorescent filters that are to be calibrated. Use at least the 20x objective, but preferably the 40x objective.
5. Navigate to an area with high contrast and focus. Then click the [Next] button to start the acquisition process.



6. Once the image has been acquired, the [Calibrate channel XY-shift] tool window opens. The shift between the individual channels is corrected manually here.
   » The visibility of the channels can be changed by activating/deactivating the 'eye' icon next to the channel name.
7. The first channel, e.g. DAPI will be used as reference. Click on a channel, e.g. FITC, to activate it and then use the arrow buttons to move the FITC image around pixel by pixel to match the structure of the underlying DAPI image.
8. Proceed with the other channels the same way.

9. When you have finished, click the [OK] button to save the changes.



» In the [Calibrations] dialog box all calibrations should have a blue tick now.

25 Cleaning the system

25.1 Cleaning the VS200 scanner

We recommend cleaning the scanner as required when you can see that it is dirty.

ATTENTION
Ingress of fluid can damage the devices.
An ingress of fluid can cause a short circuit or damage the scanner, individual components of the scanner, the loader, and connected devices.

- Before cleaning the VS200 system, disconnect it from the power supply.
- Protect the scanner, the loader, and individual components from dirt, water and ingress of fluids.
- Clean only the surface of the scanner, the loader and the individual components.
- Make sure using a dry cloth for cleaning cables and the power supplies.

1. Disconnect the scanner from the power supply.
2. Clean the following components with a lint free cleaning cloth.
   - » The surface of the scanning stage
   - » Liquid dispenser head

     Use a dry cloth for the liquid dispenser head.
3. For more pronounced marks, you can use alcohol-based disinfection wipes.
4. Clean the clamping mechanism only with a lint free cleaning cloth.
5. If the scanner or individual components were cleaned with a damp cloth, allow them to dry.

25.2 Cleaning the VS200 loader

ATTENTION
Ingress of fluid can damage the devices.
An ingress of fluid can cause a short circuit or damage the scanner, individual components of the scanner, the loader, and connected devices.

- Before cleaning the VS200 system, disconnect it from the power supply.
- Protect the scanner, the loader, and individual components from dirt, water and ingress of fluids.
- Clean only the surface of the scanner, the loader and the individual components.
- Make sure using a dry cloth for cleaning cables and the power supplies.

1. Take all of the trays out of the loader.
2. Disconnect the VS200 system from the power supply.
3. Open the door of the loader.
4. Manually move the SCARA robot arm to the right.
5. Move the tray hotel to the loading position all the way at the front.
6. Clean the following components with a lint free cleaning cloth.
   - » Left side of the tray hotel
   - » Right side of the tray hotel
   - » The bottom panel of the tray hotel
7. For more pronounced marks, you can use alcohol-based disinfection wipes.
8. Push the tray hotel as far back as it will go to the back panel.
9. Manually move the SCARA robot arm to the left.
10. Clean the gripper only with a lint free cleaning cloth.
11. If the loader or individual components were cleaned with a damp cloth, allow them to dry.

25.3 Cleaning the trays

1. Clean the trays with a lint free cleaning cloth. For more pronounced marks or remnants of immersion medium, you can lightly wet the cleaning cloth with water, 70% ethanol, or use alcohol-based disinfection wipes.

25.4 Cleaning the fluorescence light sources

✓ The X-Cite light source is not available in all countries.

Refer to the safety instructions in chapter Safety instructions for fluorescence light sources on page 16.

Observe the cleaning instructions in the manual of the light source.

1. Clean the optics only by using appropriate fluids and lens paper.
2. To clean the exterior of the unit, use a slightly dampened cloth and a simple water/ detergent solution only.

25.5 Cleaning the immersion objective

ATTENTION
Damage to the objectives and hardware
Objectives and hardware can get sticky after an immersion medium has been used. This can damage them.
- Clean the immersion objective after each use.

CAUTION
Pinching hazard from moving components inside the VS200 system
Mechanical components move around inside the VS200 system. If you attempt to make adjustments inside the system while it is switched on, your hands and fingers can get pinched or your hair and cloths can get caught.
- Make sure not to perform any software functions while you are cleaning the immersion objectives.

1. To clean lenses, remove dust by blowing them with a commercially available blower and wipe them lightly with cleaning paper. Only if they are stained by fingerprints or oils should you wipe them using cleaning paper slightly moistened with off-the-shelf dehydrated alcohol sold at store.

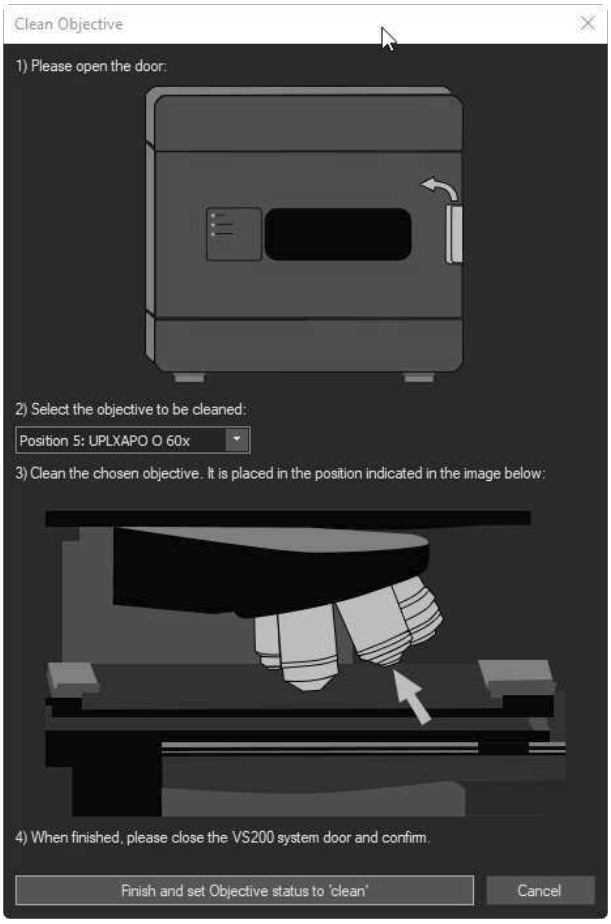
   CAUTION
   Dehydrated alcohol is highly flammable. Do not expose it to heat or flame, and do not turn off or on the power switch of various electrical apparatuses since ignition can be induced by just switching switches on and off. In addition, make sure that the room is well ventilated.

2. Do not use an organic solution to clean parts other than lenses. If a part is heavily stained, wipe it with a soft cloth slightly moistened with a diluted neutral detergent.

   Avoid cleaning lenses with the camera or the lenses removed. Doing so will cause the system to be uncalibrated. All calibrations need to be re-done.

3. Click the [Clean Objective] button on the start page of the VS200 ASW software and follow the instructions in the dialog box.
   » The objective revolver will move up, giving you more space.
   » It is not necessary to unmount an objective to clean it.

4. In the [Select the objective to be cleaned] list select the objective to be cleaned. If you have more than one immersion objective, also select the other objectives in the list.



5. To clean the objective use lens cleaning paper.
6. After cleaning the objective click on the [Finish and set Objective status to 'clean'] button.

25.6 Cleaning the liquid dispenser

1. Before starting the maintenance, switch off the scanner and unplug the power cable.
2. On the liquid dispenser head:
   » Use tissue with ethanol and gently wipe to clean any oil residues from the liquid outlet.
3. Check if oil is accumulating on the condenser top lens:
   » If the dispenser is not being used for a long time, oil inside might turn viscous and affects the closing of the valve. As a result, oil leaks from the liquid outlet and will accumulate on the condenser top lens.

» If oil is found on the top lens, use lens tissue and ethanol to clean it.

» Report to your local service immediately.

26 Installing additional software

26.1 OlyVIA

To install OlyVIA go to D:\\EVIDENT_SERVICE_ONLY_DO_NOT_DELETE\SetupOlyvia and execute the [setup.exe].

26.2 VS200 ASW Desktop

VS200 ASW and VS200 ASW DT cannot be installed on the same customer PC.

The installation requires a VS200 ASW DT licence key during the installation.

To install VS200 ASW DT on a different PC copy the setup files to the desired PC and execute the setup.exe. The setup files can be found on the VS200 ASW system PC under the following link: D:\EVIDENT_SERVICE_ONLY_DO_NOT_DELETE\SetupMain

26.3 NetImage Server SQL (NIS-SQL) and Webinterface

The NetImageServer SQL can be installed by Evident service technicians only.

Also the web interface to utilize OlyVIAweb (HTML5-based webviewer) is installed and configured by Evident service technicians only.

27 VS200 Speckle Illumination Acquisition (SILA)

The units described below must be assembled and adjusted by Evident. If these units are assembled or adjusted by the customer, the operations are not ensured.

27.1 Safety

Before installing and using the VS200 system with SILA make sure to read and follow the safety instructions in the safety chapters of this manual. See Safety on page 11.

When using the VS200 system with SILA, make sure to be familiar with the user instructions for all connected devices. For safe operation of the laser combiner and the SILA hardware refer to the respective manuals.

27.2 Intended use

The VS200 Speckle Illumination Acquisition (SILA) optical sectioning device is an optical microscope accessory designed for general laboratory use only. It is both a hardware and software solution, producing high contrast and blur-free images of thicker specimens.

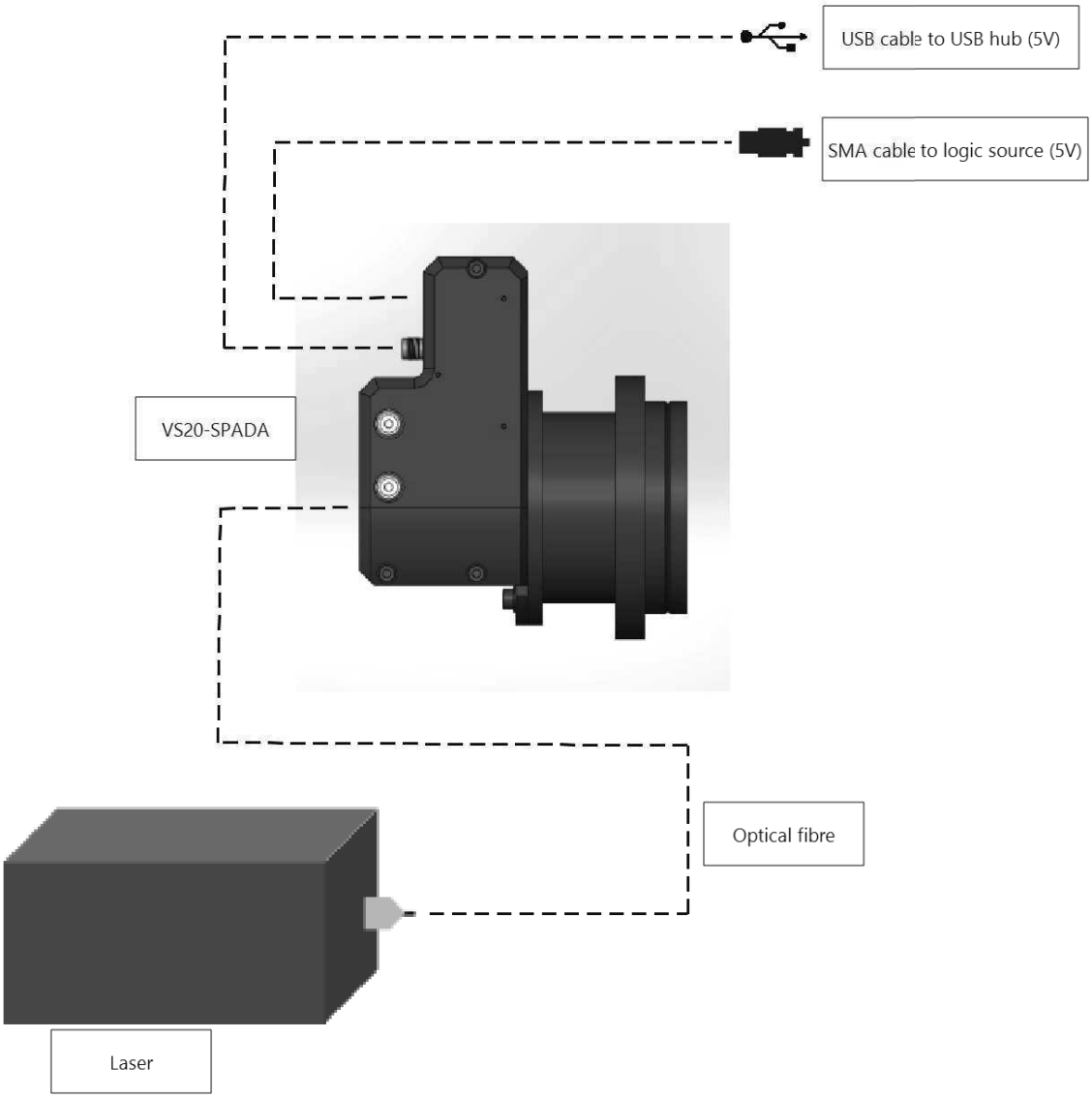
It's main components are a laser combiner which consists of up to 6 laser lines, and a SILA hardware which can produce speckle illumination. The SILA solution can only be used with the VS200 slide scanner only.

27.3 Scope of supply

When ordering a VS200 system with SILA the following items are supplied, in addition to the base unit:

VS20-SPADA	VS200 hardware for SILA
VS20-LASER or C-FLEX C6	Laser combiner 4 lines or Laser combiner 6 lines
VS20-LAS-LEMO	VS20-LAS-LEMO consists of the LEMO cable for speckle illumination, power extension cable, screws and laser class labels
VS20-LAS-PWSUP	Laser power supply
VS20-TRIGGER	PCIe trigger card for VS20-SPADA and laser combiner

27.4 System diagram



27.5 Specifications

		VS200 SLIDEVIEW Slide Scanner with Fluorescence
Intended specimen	Observable specimen	Glass slide with coverslip
	Thickness of coverslip	0.12 – 0.17 mm
	Thickness of specimen	Up to 0.5 mm
Imaging	Observation methods	Fluorescence - optical sectioning with Speckle Illumination
	Magnification	4x, 10x, 20x, 40x, 60x and 100x (incl. selected oil immersion and silicon oil immersion objectives)
	Sectioning thickness	1 to 10 x depth of field of the objective in use
	Exposure	Minimum: 50 ms
	Scan Time	Approx 14 minutes (20x objective scan area 15 mm x 15 mm, 4 channels, 50 ms exposure each)
Light Source	Wavelengths	Up to 6 laser lines, customizable wavelengths among a selection ranging from 375 to 785 nm
	Optical fiber	Single mode fiber (connector: FC/APC)
	Laser combiner	Laser Power: 100mW AC adapter: 15 VDC, 7A
SILA Hardware	Activation time	Rise time: 10 ms Fall time: 20 ms
	Maximum modulation frequency	10 ms
	Lifetime	20,000 hours
	Dimensions	H/W/D: 72.0 / 98.7 / 88.5 mm Weight: 520 g
	Operating environment	Temperature: 12 – 28 °C Humidity: up to 80% (31°C) Altitude: 2000 m
	Power	5 VDC, 0.5 A
	TTL control input	SMA connector Threshold: 3.0 V

27.6 Unboxing

The units described below must be assembled and adjusted by Evident. If these units are assembled or adjusted by the customer, the operations are not ensured.

VS20-SPADA

1. The VS20-SPADA is packed in two layers of boxes. After reaching the second box, take out the SMA-SMA signal cable (gold ends) and USB power cable from the box. They are located on top of the foam.

2. Take the foam out of the box.
3. Take the ESD bag containing the VS20-SPADA main unit out of the box.
4. Put latex gloves on.
5. Open the ESD bag and take out the VS20-SPADA main unit. Handle them with care

VS20-LASER or C-FLEX C6

1. Components of VS20-LASER or C-FLEX C6 are individually packed and placed securely in foam that is perfectly cut to shape. First remove the boxes containing the optical fiber and accessories. Accessories required for installation are: TTL signal cable (thick cable with green end), remote

interlock jumper, keys and USB-B to USB-A data cable.

2. Remove the top layer of the foam and take out the laser combiner.

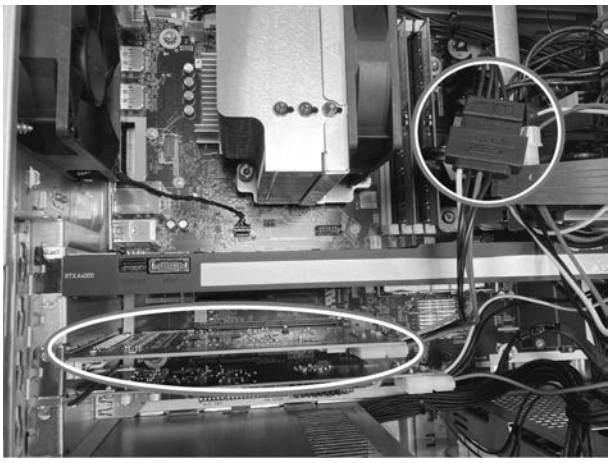
27.7 Installing the SILA components

The units described below must be assembled and adjusted by Evident. If these units are assembled or adjusted by the customer, the operations are not ensured.

This step can be started after fully assembling the hardware of VS200 system with fluorescence parts as described in the chapters Mounting fluorescence components on page 53 and Assembly of the housing for the VS200 scanner on page 81, excluding the top and back panel of the base unit housing. In case a dual lamp housing is used, it also needs to be assembled before this step. For this, refer to the dual lamp housing manual.

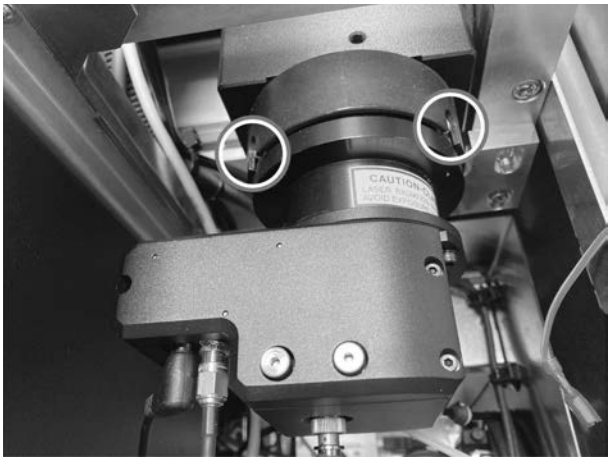
27.7.1 VS20-TRIGGER

1. Remove the cover of the VS200 PC.
2. Plug in VS20-TRIGGER into an empty PCIe slot. Remember to also connect the power supply. The power extension cable for the PCIe-trigger card is included in the VS20-LAS-LEMO package.

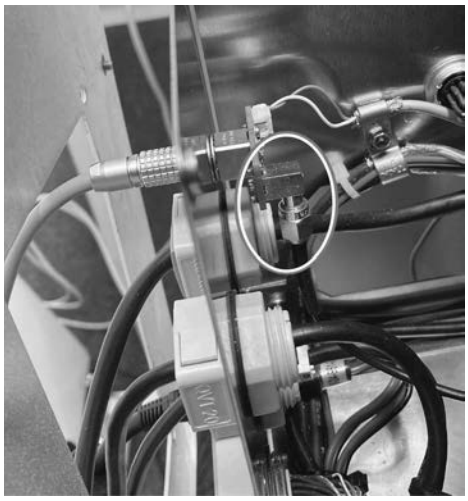


27.7.2 VS20-SPADA

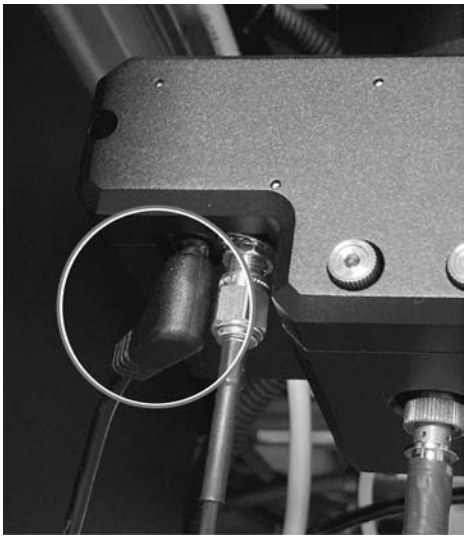
1. Mount the main unit of the VS20-SPADA to the IX3-RFALFE or connect it to the (front or lateral) port of the dual lamp housing. For more information on the dual lamp installation refer to the installation manual for the dual lamp housing. Tighten the fixation screws using a 3 mm hex key.

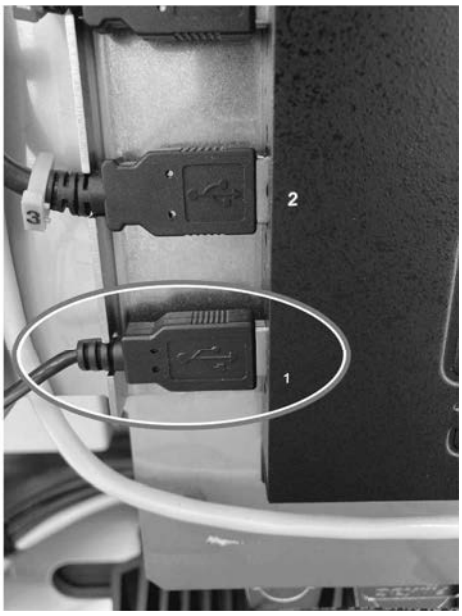


2. Roll/arrange the SMA-SMA signal cable inside the device and connect it to the VS20-SPADA (left picture) and to the back connector panel of the base unit (right picture).



3. Run the power USB cable inside the device and connect it to the VS20-SPADA and to the USB-Hub Port 1 at the back of the base unit.





27.7.3 Laser combiners: VS20-LASER or C-FLEX C6

1. Place the VS20-LASER or the C-FLEX C6 on a table in such a way, that all control elements are accessible and the optical fiber can be run without being bent. The key switch and indicator lights should face the front, the other ports should face the back. If applicable, fix the laser combiner to the table, referring to the main C-FLEX manual.

2. Place the remote interlock jumper, a 2-pin molex jumper, into the remote interlock socket.

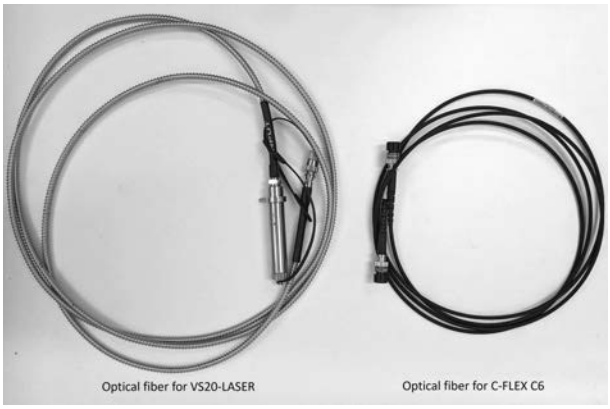
3. Connect the laser combiner with the laser power supply.

4. Connect the USB type B cable to the laser combiner and to the PC. See USB position to use in chapter Cabling on page 82.
5. Connect the TTL signal cable to the laser combiner and to VS20-TRIGGER installed on the PC.

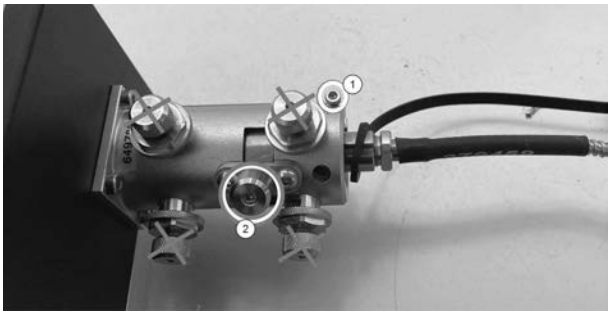
6. Connect the silver VS20-LAS-LEMO cable to the laser combiner and to the back connector panel of the VS200 system.

27.7.4 Mounting the optical fibre

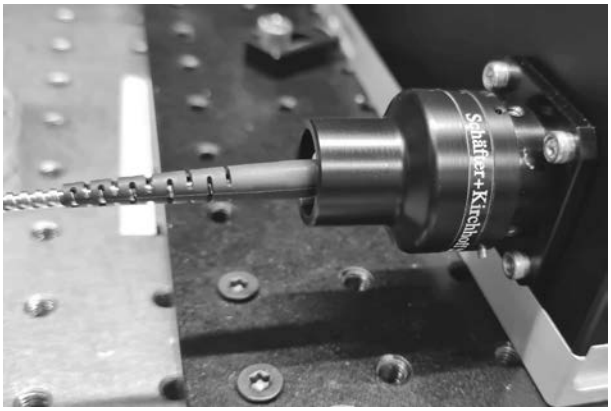
1. Partially mount the back panel of the VS200, using only the fixing screws on the bottom part to enable easy installation of the optical fibre.
2. Carefully unpack the optical fibre. Avoid applying any pressure to it, especially bending it.



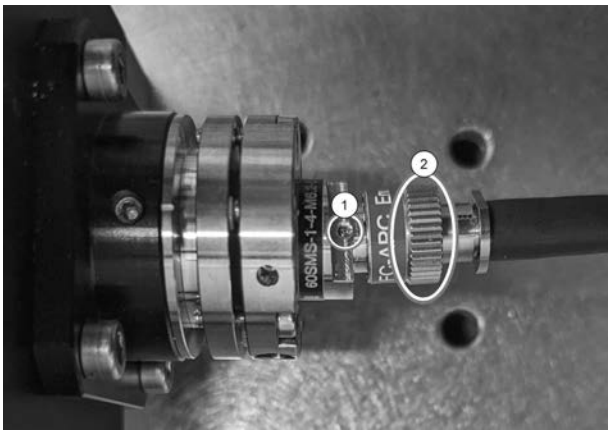
3. Attach the optical fibre to the laser combiner coupler.
   - » Procedure for VS20-LASER combiner: Remove the small locking screw (1) from the coupler (hex key 2 mm), push locking button to the coupler (2) and insert the optical fiber connector in the coupler till the end. Reattach the locking screw (1). Do NOT touch the other screws as this will misalign the lasers.



   - » Procedure for C-FLEX C6 combiner: Unscrew the 3 screws to remove the protection cap.



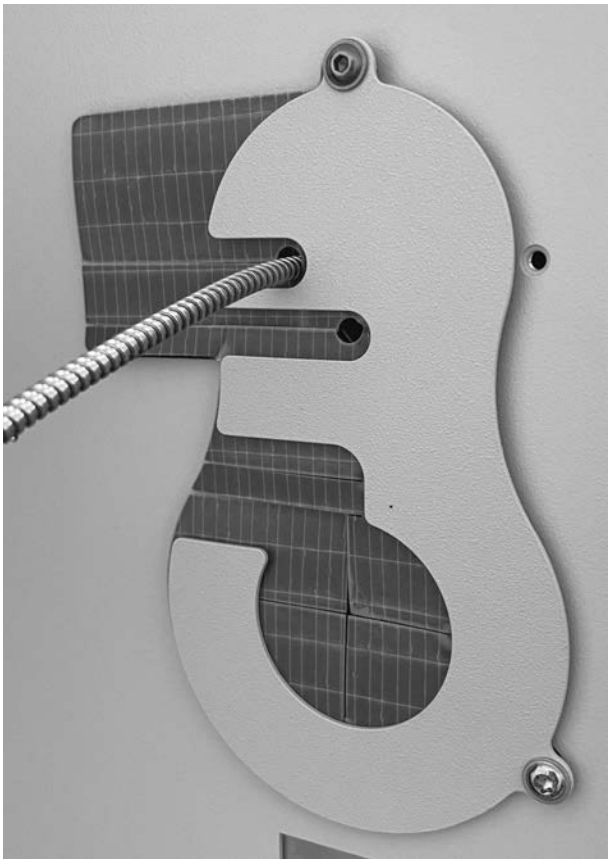
Loosen the small locking screw (1) from the coupler (screw driver provided by cobolt) - do not remove it from the position completely to avoid its loss. Insert the optical fiber connector in the coupler till the end. Tight the optical fiber lock (2) and at last tight the locking screw (1).



Run the protection cap along the optical fiber and mount it back to the original position using the 3 screws.

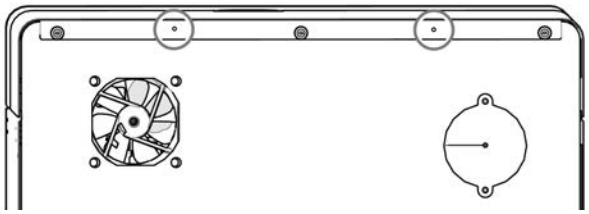
4. Run the optical fibre through the opening in the back panel of the VS200, and then attach the optical fibre to the VS20-SPADA.

5. Place the back panel against the backside of the system and tighten the four knurled hex screws. Do it carefully to avoid any impact on the optical fibre.
6. Place the protective cover over the hole. Do it carefully to avoid any impact on the optical fibre.



27.7.5 Top back panel

1. Mount and tighten the 3 knurled hex screws of the top back panel. See Left, right, back and top panels on page 81.
2. The top back panel of VS200-BU-V2, VS200-BU-L-V2 comes with two additional 4 mm hex screws which can be secured to the back panel. Tighten these screws. The screws are included in the VS20-LAS-LEMO package.



27.7.6 Top front panel

1. Mount the top front panel. See Left, right, back and top panels on page 81.
2. The top front panel of the VS200-BU-V2, VS200-BU-L-V2 comes with two additional hex screws (size 2 mm hex key) connecting to the black tamper protection plate. Tighten these screws. The screws are included in the VS20-LAS-LEMO package.

27.7.7 U-FF filter cube containing multi-band dichroic mirror

The VS200 system with SILA uses a multi-band dichroic mirror which is compatible with lasers, for example 89402bs from Chroma. Note that an excitation filter is not required for SILA. Individual emission filters are recommended.

1. Open the door of the VS200 scanner.
2. Slide the door of the IX3-RFACA to the left to insert the filter cube.

3. Insert a filter cube into the correct position (position 2) as shown in the image. If you are installing the C-FLEX C6 laser combiner, it might be necessary to add more than one filer cube.

The correct position is printed inside the IX3-RFACA and is hard to read as the IX3-RFACA is mounted upside down. If you cannot read it make sure that the filter is placed between the position index 7 and 8 as shown in the image below.

4. After the cube is inserted slide the door of the IX3-RFACA back to the right.

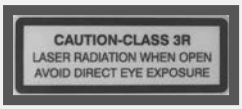
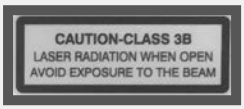
27.7.8 Metal plate (loader only)

1. Unpack the metal plate and remove the protecting film.
2. Open the loader door and stick the metal plate to the internal side of the glass opening.

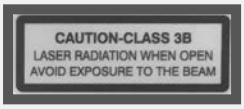
After mounting this component securely, the system is now completely closed and regarded as a Class 1 laser equipment.

27.8 Placing the laser class safety labels

Position of safety labels for the VS200 system with SILA using the VS20-LASER and C-FLEX C6 laser combiner. The labels are included in the VS20-LAS-LEMO package.

	Laser class safety label - 3R (for VS20-LASER)
	For the VS20-LASER: Laser class safety label 3R to be placed at the metal frame under the top front panel of the base unit.
	Laser safety class label - 3B (for C-FLEX C6 laser combiner)
	For the C-FLEX C6 laser combiner: Laser safety class label 3B to be placed at the metal frame under the top front panel of the base unit.

VS20-SPADA

	On the VS20-SPADA unit Laser class safety label - 3B for the VS20-LASER and for the C-FLEX C6 laser combiner.
	To be placed at the VS20-SPADA unit when mounting it to the IX3-RFALFE.

	To be placed above the opening where the optical fiber is connected to VS20-SPADA, at the upper right corner of the back panel.

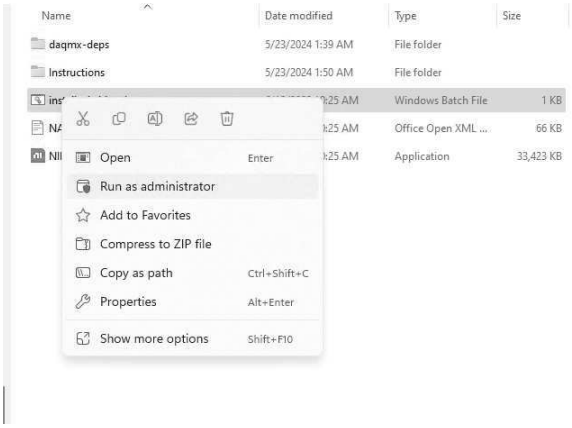
27.9 National Instruments (NI) software installation

The units described below must be assembled and adjusted by Evident. If these units are assembled or adjusted by the customer, the operations are not ensured.

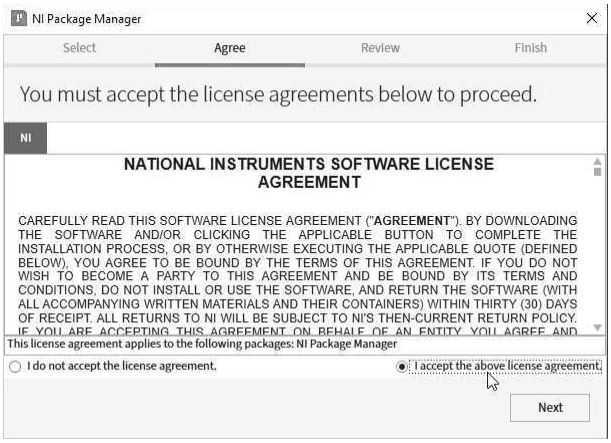
This software is required to control the lasers during SILA acquisition.

✓ Start this step after you have completed the VS200 ASW installation and adjusted the COM ports of the system. See Adjusting WINDOWS COM ports on page 108.

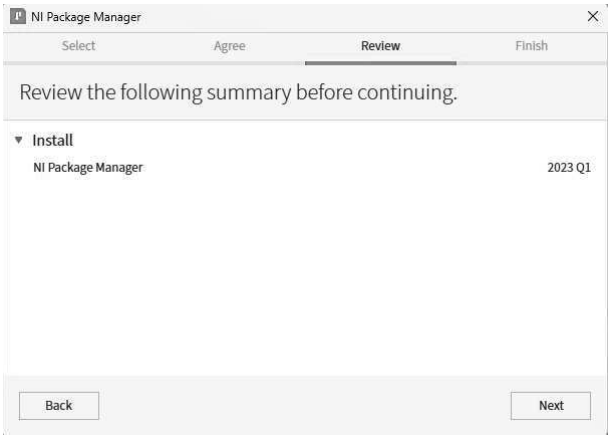
1. The driver to install the National Instrument card is included in the setup.iso-file (OLY-MNX-<buildnumber>.iso). To access it double click on the OLY-MNX-<buildnumber>.iso.
2. Right click on the Windows batch file install-ni-driver.bat and select [Run as administrator] from the drop down menu.



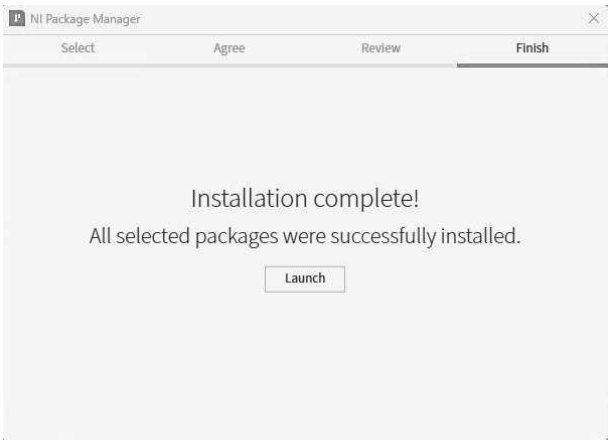
3. Accept the license agreement.



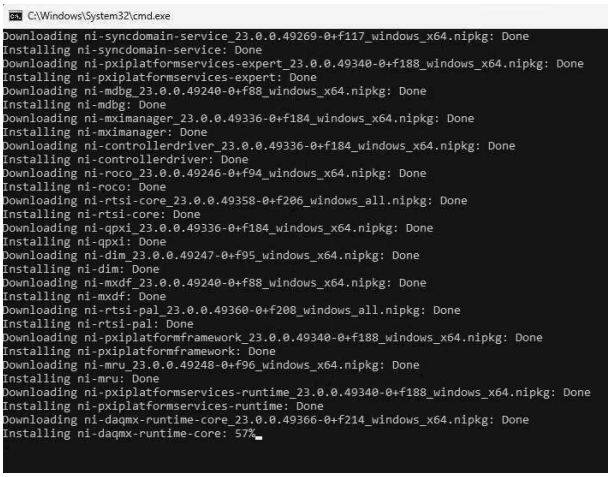
4. Review the installation package and click [Next].



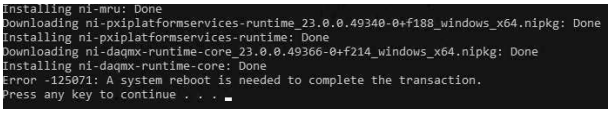
5. After the installation of the [NI Package manager] software, close the dialog box.



6. The installation of the NI-daqmx will start automatically. You can monitor it in the Windows dialog box.



7. When the installation finishes press any key and reboot the system. The installation will be completed after reboot.


Installing ni-mru: Done
Downloading ni-pxiplatformservices-runtime_23.0.0.49340-0+f188_windows_x64.nipkg: Done
Installing ni-pxiplatformservices-runtime: Done
Downloading ni-daqmx-runtime-core_23.0.0.49366-0+f214_windows_x64.nipkg: Done
Installing ni-daqmx-runtime-core: Done
Error -125071: A system reboot is needed to complete the transaction.
Press any key to continue . . .

27.10 Adjusting COM ports for the lasers using Cobolt Monitoring tool and Device Manager

The units described below must be assembled and adjusted by Evident. If these units are assembled or adjusted by the customer, the operations are not ensured.

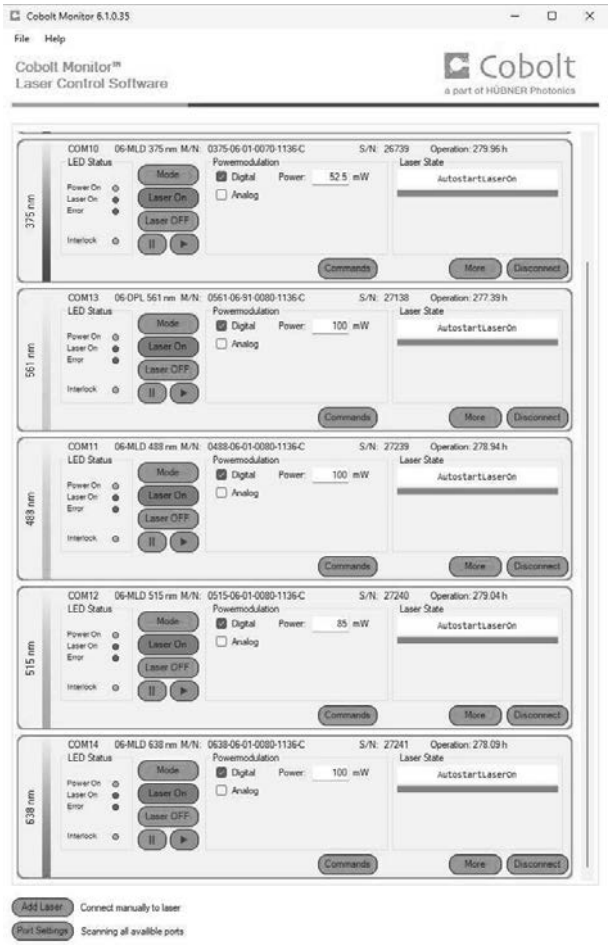
The Cobolt Monitoring tool is required to check the functionality of the lasers and to read out the COM ports used.

The Cobolt Monitoring tool and our VS200 ASW software cannot be opened at the same time as they will occupy the same ports.

1. The Cobolt Monitoring tool can be found in the following location: https://hubner-photonics.com/downloads/

   The latest version which works with both VS20-LASER and C-FLEX C6 is CoboltMonitor 6.1.4.0.

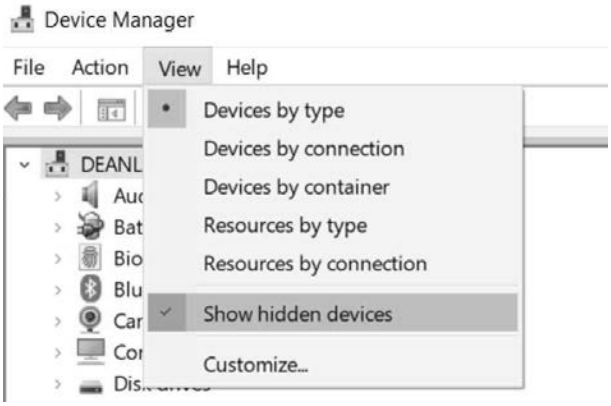
   » The available laser lines and the COM ports they occupy are displayed in the following image. The port numbers may differ with each PC.



In the VS200 ASW software each laser line is assigned to a specific COM port to ensure successful connection. The lasers must be assigned to COM ports according to their wavelength, starting from COM10 for the shorter wavelength (e.g. 375 nm) until a maximum of COM15 for the longest wavelength (e.g. 730 nm). An example of a set up with 5 laser lines is illustrated in the screenshot above.

If the Cobolt Monitor shows a mismatch in the COM port order, they need to be readjusted using the Windows [Device Manager] dialog box.

1. Take a screenshot of the Cobolt monitor, or take a note to which COM port the lasers are currently assigned.
2. Close the Cobolt monitor and open the MS Windows [Device Manager] dialog box.
3. Check in the [Device Manager] dialog box if COM ports 10 to 15 are already occupied. If they are occupied by a hidden device, change these devices to a higher port number to free up these COM ports for the lasers.
4. To do so, select in the menu of the [Device Manager] dialog box the [View] > [Show hidden devices] entry.

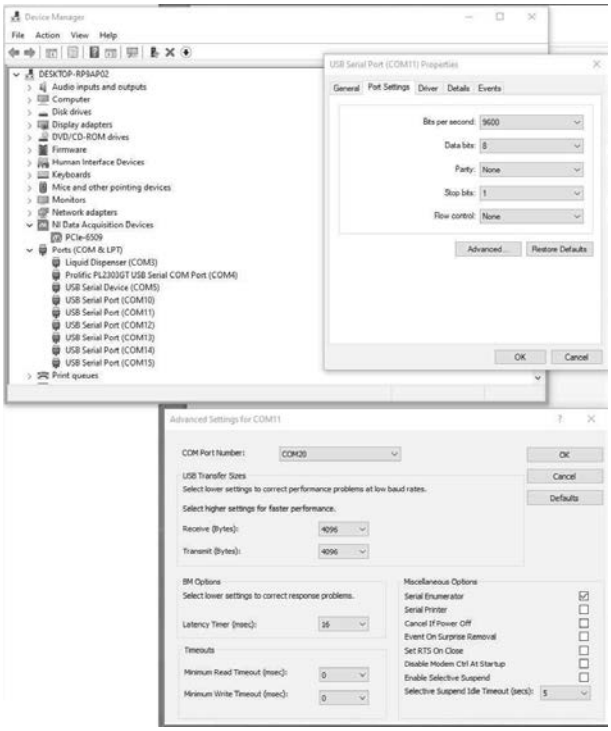


» For example if [COM11] is occupied, right click on [COM11] and select the [Properties] entry in the context menu.

» Click [Port Settings] then click [Advanced].

» Change the COM port number to a higher value, e.g. 20 and click [OK].

» Repeat this step until all occupied COM ports from 10 to 15 are free.



5. Change the port numbers of the laser lines. For example, 488 laser line should occupy COM11 but it is at COM12. Right click on COM12 and select the [Properties] entry in the context menu.
6. Click the [Advanced] button. Change the COM port number to 11 and click [OK].
7. When all the lasers are correctly assigned, close the [Device Manager] dialog box and restart the PC.
8. Start the Cobolt monitoring tool and check that the lasers are correctly assigned.
9. Proceed with the VS200 device configuration. See VS200 device configuration on page 112.

27.11 User operation of VS200 system with SILA

When operating a correctly installed, faultless VS200 system with SILA , no laser radiation above Class 1 levels can exit the system. This chapter describes the actions that are allowed when operating the VS200 system by the user and lists restrictions.

All hardware has to be installed properly by trained service personnel before the system can be operated by a user. In normal usage, the user can operate the system without being exposed to the laser beam. It is possible to have full control of the various devices via software. When operating the VS200 system with SILA as intended by a user the VS200 system with SILA is treated as a Laser Safety Class 1 equipment.

Permitted actions for a user for normal operation	
Permitted action	**Remarks**
Change of trays from base unit or loader	Safety shutter is closed automatically
Live observation or image acquisition via software	Access to laser radiation above Class 1 levels is blocked
Change of software settings and control of microscope components (such as objectives or filters) through software, as long as no hardware is being dismounted and no tools have to be used for access	Access to laser radiation above Class 1 levels is blocked or safety shutter closes automatically

Permitted actions for a user for maintenance	
Permitted action	**Remarks**
Cleaning of the imaging system where accessible without tools	
Cleaning of optics of the imaging system where accessible without tools	Safety shutter is closed automatically
Change of microscope components, as long as no tools have to be used in the process. For example, exchanging objectives on the nosepiece or filter cubes in the fluorescence turret.	Safety shutter is closed automatically

Strictly prohibited actions for a user

» Any removal, change or modification of the system where tools are required such as: Removing/changing the camera, remove any part of the VS200 casing, detaching optical fibers.

Actions that are only to be carried out by service personnel, described in the Installation Manual or Repair Manual like:

» Set up and connection of the VS200 SILA components.

» Alignment of lasers.

Starting operation

1. Open the mechanical safety shutter by moving the slider which is located on the back of VS20-LASER or on the back of the C-FLEX C6 laser combiners.

2. Insert the key and rotate it to the ON position (key in horizontal position). If it is already in the ON position, turn it to OFF and then ON again. Once the key is turned to the ON position the lasers will automatically start up. Wait at least 3 minutes for the lasers to stabilize.

3. Verify that all status LEDs are green. Now the device hardware is ready for operation.
4. Start the VS200 ASW software. Information on how to operate the software can be found in the online help of the VS200 ASW software.

To protect your specimen from photo bleaching, switch off the live view in the software whenever suitable, even if you interrupt observation only for a short time.

Ending operation

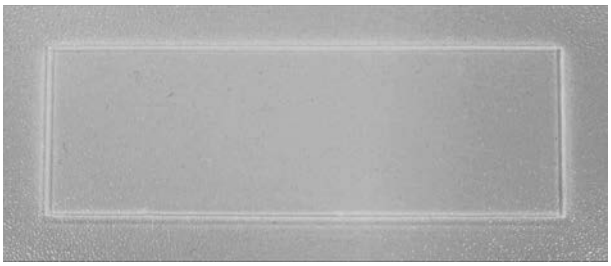
1. To finish the operation, close the VS200 ASW software. Rotate the key to the OFF position.
2. Unplug the power supply.

27.12 SILA alignment check

The units described below must be assembled and adjusted by Evident. If these units are assembled or adjusted by the customer, the operations are not ensured.

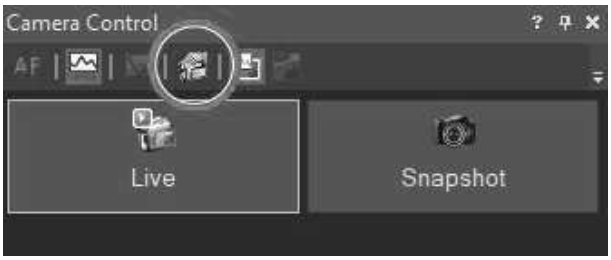
» Autofluorescence plastic slide (for example part number 92001 from Chroma Technology Corp)

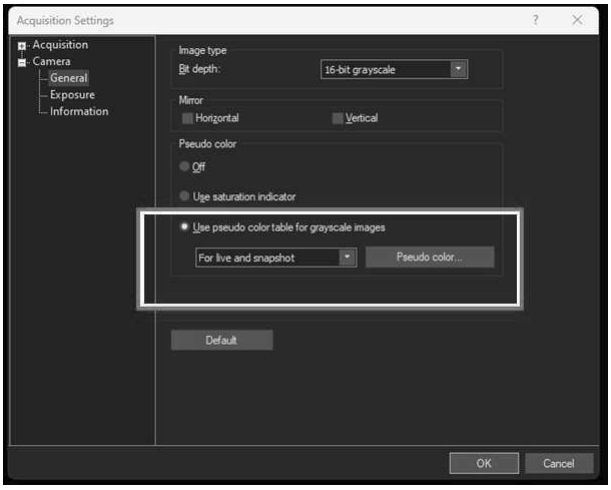
1. Put a green autofluorescence plastic slide (for example part number 92001 from Chroma Technology Corp) onto an empty tray and choose this tray for calibration.



2. In the VS200 ASW software, go to the [Manual control] layout.
   » When you are in the [Scan] layout, you can use the [Additional layouts] button to switch the layout.
3. Select the 20x objective. Start with 488 laser line, move to the green Chroma slide and select the [FITC] observation.
4. In the [Camera Control] tool window, open the [Acquisition Settings] > [Camera] > [General] dialog box.

   In the [Pseudo color] group, select the [Use pseudo color table for grayscale images] option. Select the [For live and snapshot] entry from the list.





5. Load the [Rainbow-BlackToRed] pseudo color table to better visualize the laser beam center.

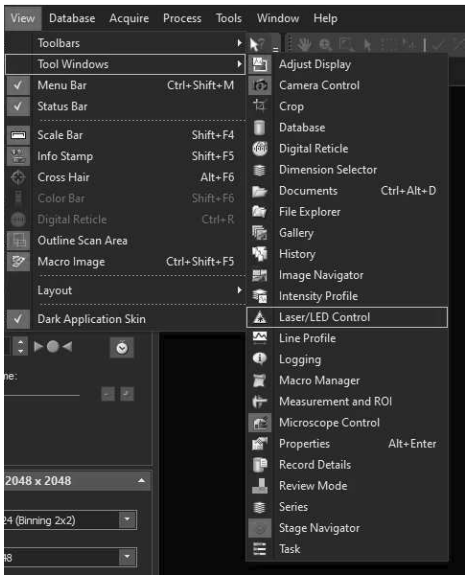
   To do so, click the [Pseudo color] button to open the [Pseudo Color] dialog box.

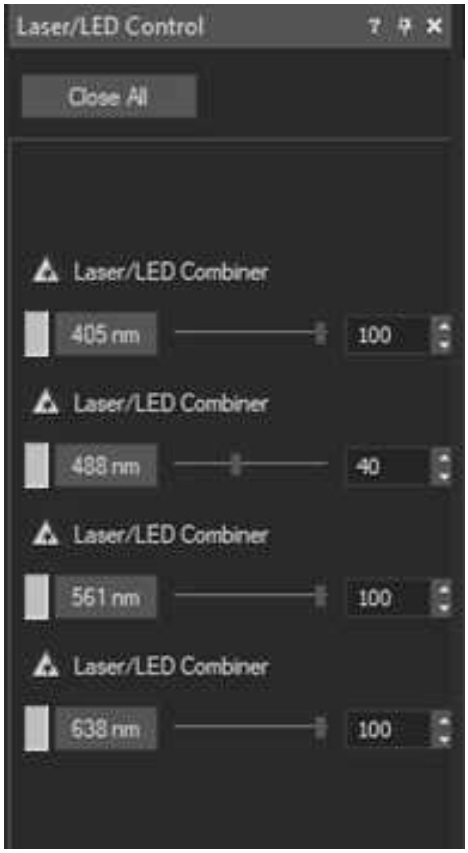
   In the [Pseudo Color] dialog box, click the [Load Color Table] button. Select the [Rainbow-BlackToRed] entry and click the [Load] button.

   Close all open dialog boxes by clicking [OK].

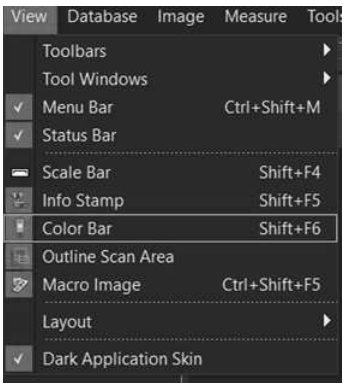
6. Open the [Laser/LED Control] tool window. To do so, use the [View] > [Tool Windows] > [Laser/LED Control] command.

   In the [Laser/LED Control] tool window, reduce the laser intensity of the 488 nm laser to 40%.

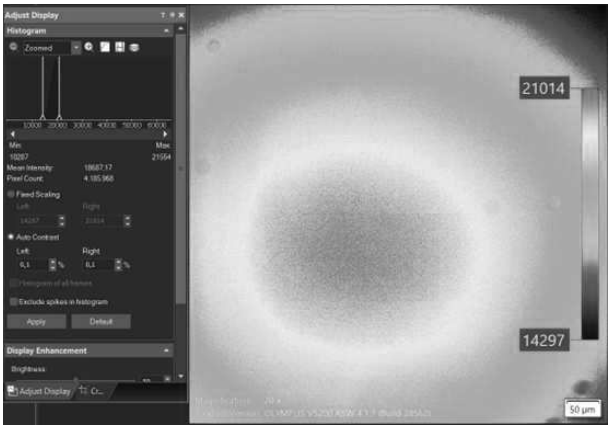




7. Adjust the exposure time to minimum and start the live mode.
8. Open the [Adjust Display] tool window. Use the [Histogram] group to make sure that the image is not over or undersaturated. Adjust the laser intensity and/or exposure time if required.
9. In the [View] menu, select the [Color Bar] entry to show the maximum and minimum intensity of the image.



10. Make sure the display limits are set to [Left:] 0.1%, [Right:] 0.1% and click the [Apply] button.



11. Divide the lower intensity value with the higher one to find out the percentage difference in intensity between the center and edge of the image.

    For example: 14297/21014 * 100% = 68%.

    a. If the value is within 70±10%, proceed with imaging.

    b. If the difference is significantly smaller than 70%, contact your Evident technical service representative.

12. After the alignment checking is done, switch off the pseudo color mode to get back to the normal image display mode.

    In the [Camera Control] tool window, open the [Acquisition Settings] > [Camera] > [General] dialog box.

    In the [Pseudo color] group, select the [Off] option. Close all open dialog boxes.

27.13 Troubleshooting of the VS200 system with SILA

The units described below must be assembled and adjusted by Evident. If these units are assembled or adjusted by the customer, the operations are not ensured.

27.13.1 Live image completely dark

If the live image is completely dark, do the following checks.

» Check if the laser combiner and laser key is switched on, and the mechanical interlock is set to open. See Starting operation on page 254.

» Check if the dichroic mirror is placed inside the fluorescent mirror turret. Then, check in the [Device Settings] dialog box and in the [Device Customization] dialog box if the dichroic mirror and emission filter is in the position specified by the observation method.

» Check if the base unit door is completely closed.

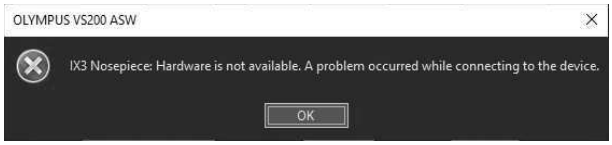
» Close the VS200 ASW software. Open the Cobolt monitoring tool and the [Device Manager] dialog box to check if the COM ports are assigned correctly. If not, proceed to adjust them. See Adjusting COM ports for the lasers using Cobolt Monitoring tool and Device Manager on page 250.

» Shut down the PC. Check if the TTL cable is securely connected to the laser combiner. See Laser combiners: VS20-LASER or C-FLEX C6 on page 237.

» Check if the power supply is connected to the PC. See VS20-TRIGGER on page 235.

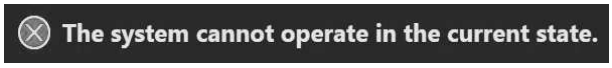
28 Troubleshooting

28.1 Hardware not available



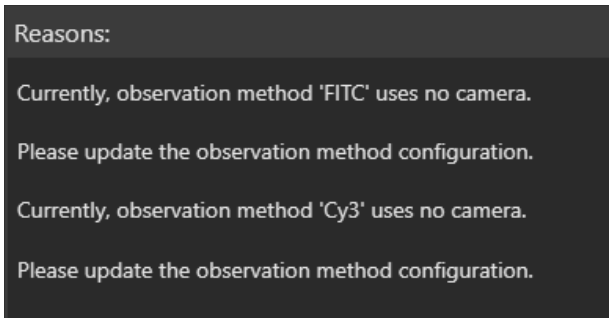
If the [IX3 Nosepiece: Hardware is not available] error message appears, do the following:

1. Close the VS200 ASW software.
2. Switch off the VS200 system!
3. Check cabling of IX3-RFACA.



28.2 "No camera" error

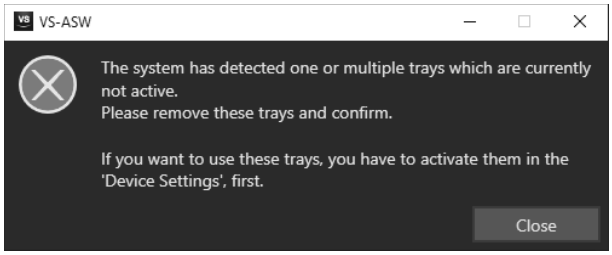
If you see the following error messages after switching to the monochrome camera you need to check whether the correct camera is assigned in the observation methods which use a monochrome camera. See Manual device configuration on page 119.

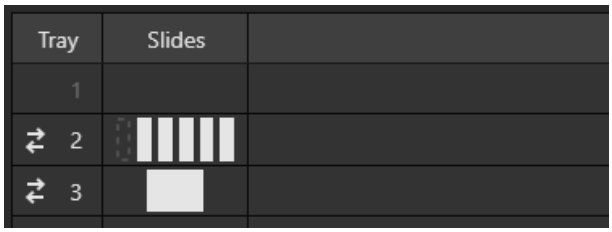


» If you are using an ORCA camera make sure that it is switched on.

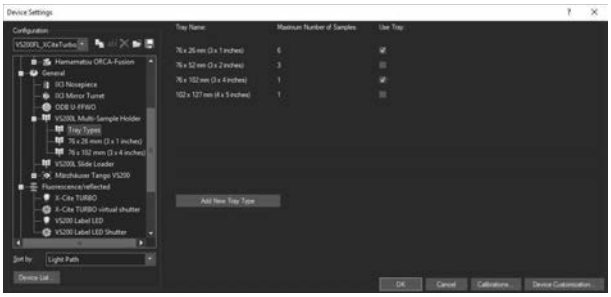
28.3 Tray not active

If you see the error message below you possibly inserted a new tray type without having it set up correctly in the VS200 ASW software first.



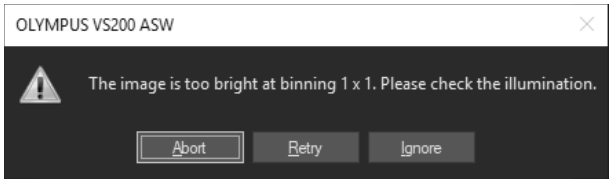


1. Remove the tray and switch to the [Manual Control] layout.
2. Use the [Acquire] > [Devices] > [Device Settings] command to open the [Device Settings] dialog box.
3. Select the [VS200L Multi-Sample Holder] > [Tray Types] entry in the tree view.
4. Select the check box [Use Tray] next to the tray type you want to use.



28.4 Image too bright

If you see the following error message please adjust VS200 LED lamp voltages. See ORCA camera adjustments on page 121.



28.5 Setting the Koehler illumination

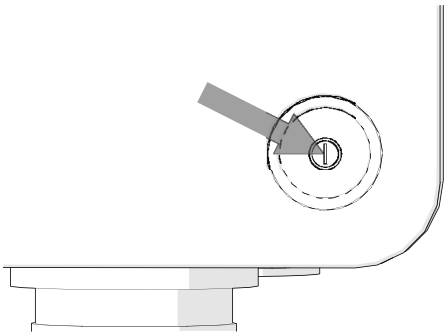
The units described below must be assembled and adjusted by Evident. If these units are assembled or adjusted by the customer, the operations are not ensured.

This chapter describes how to set the Koehler illumination.

The condenser is centered ex works in XYZ direction.

» Hex key (size 2.5mm and 3mm with ball end)

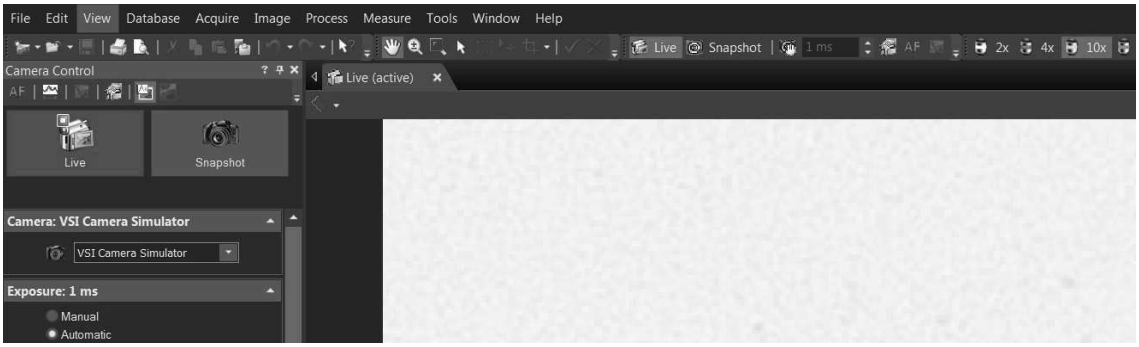
1. Use the main power switch to switch on the VS200 system.



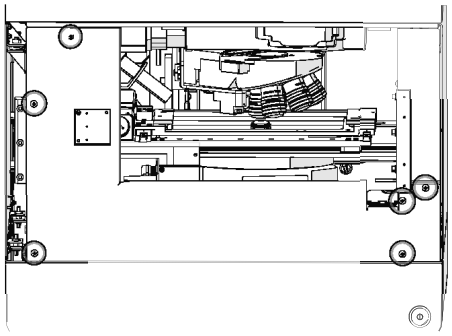
CAUTION
Pinching hazard when initializing the VS200 system
Switching on the VS200 scanner and starting the VS200 ASW software will initialize all components. As a result, various components begin to move.
Gaps open and close when the stage moves. Hands and fingers can get pinched.

- Make sure that you are not within the stage's range of movement when it is moving.
- Try never to put your hands or fingers into any gaps.

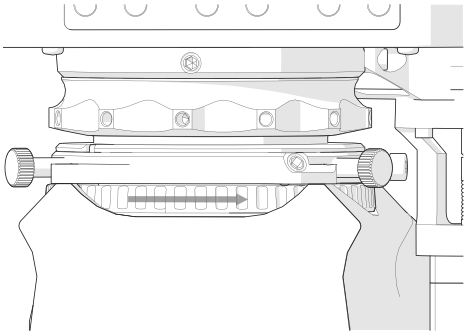
2. Start the VS200 ASW software.
3. Take a tray that has a slide in position 3.
4. On your software's start page, click the [Exchange Trays] button to load the tray.
5. Use the [Select Slide for Calibration] function to load the slide.
6. Make sure that the 2x objective is selected.
7. Use the VS200 ASW software to start the live image. To do so, go to the [Manual Control] layout.
8. In the [Camera Control] tool window, click the [Live] button to start the live image. Alternatively, you can also use the [F7] key.



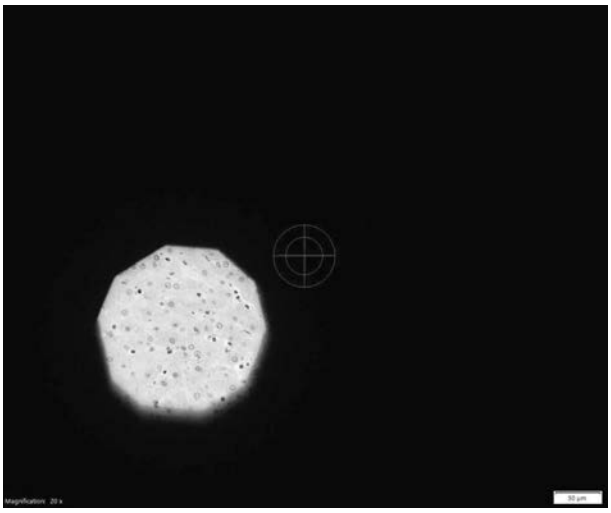
9. Carry out an autofocus.
10. Change to the objective with the magnification 10x or 20x.
11. Carry out an autofocus again.
12. Open the door of the VS200 scanner.
13. Remove the lower tamper protection plate. To do so, loosen the 6 hex screws (size 2.5 mm hex key) indicated in the figure.



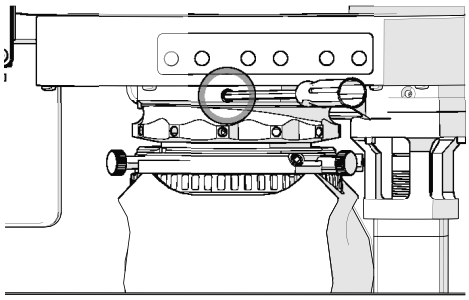
14. Turn the adjustment ring indicated in the figure to close the field stop as far as possible.



» You should now see a bright spot somewhere in the live image. The spot may appear polygonal (10 edges).
Display a cross hair in the live image to help with centering. To do so, use the [View] > [Cross Hair] command in the VS200 ASW software. Alternatively, you can also use the [Alt + F6] keystroke.

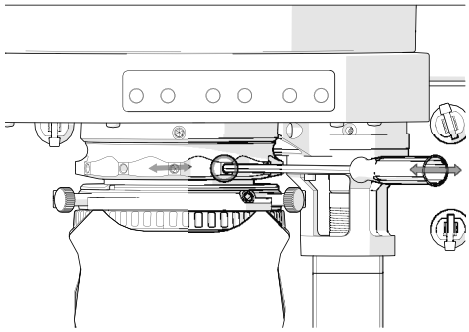


15. Release the field stop's focus setting. To do so, loosen the hex screw (size 3 mm hex key) indicated in the figure.

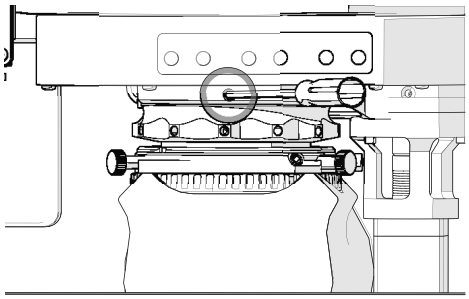


16. Focus the field stop. This can be done by hand. Alternatively, you can insert the hex key into one of the holes indicated in the figure. Move the hex key to the right and to the left to adjust the focus ring.

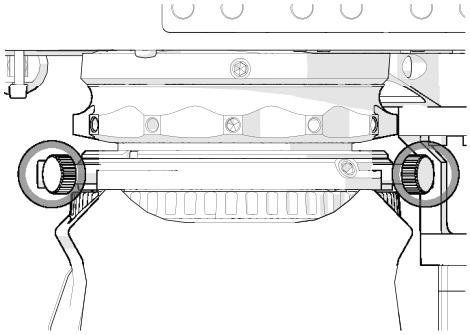
    Set the focus so that the edges of the closed field stop are sharp.

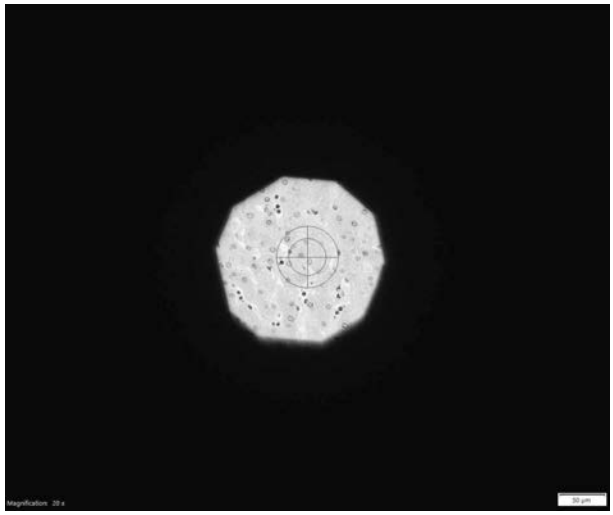


17. Tighten the field stop's focus setting. To do so, use the hex screw (size 3 mm hex key) indicated in the figure.

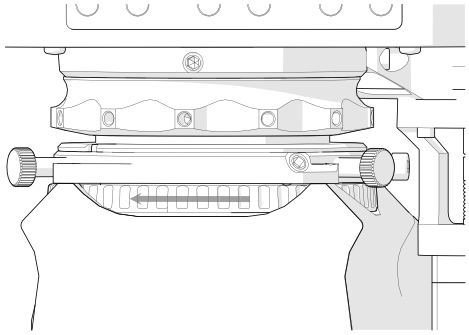


18. Loosen the 2 hex screws (size 3 mm hex key) indicated in the figure. The screws should only have been screwed hand tight.
19. Use the two adjustment screws indicated in the figure to align the bright spot in the center of the cross hair.





20. Open the field stop completely. To do so, turn the adjustment ring shown in the figure to the left as far as possible.



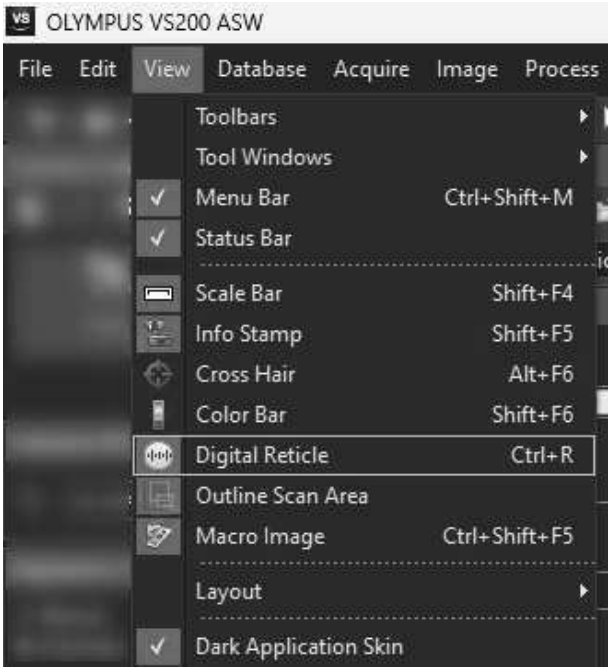
21. Mount the black tamper protection plate using the 6 screws and close the door.
22. On your VS200 ASW software's start page, click the [Exchange Trays] button to remove the tray. Confirm that you exchanged the tray so the flap will close again.
23. Close the VS200 ASW software and wait until the VS200 scanner has reached its end positions.
24. Use the main power switch to switch off the VS200 system.

28.6 Color camera rotation adjustment

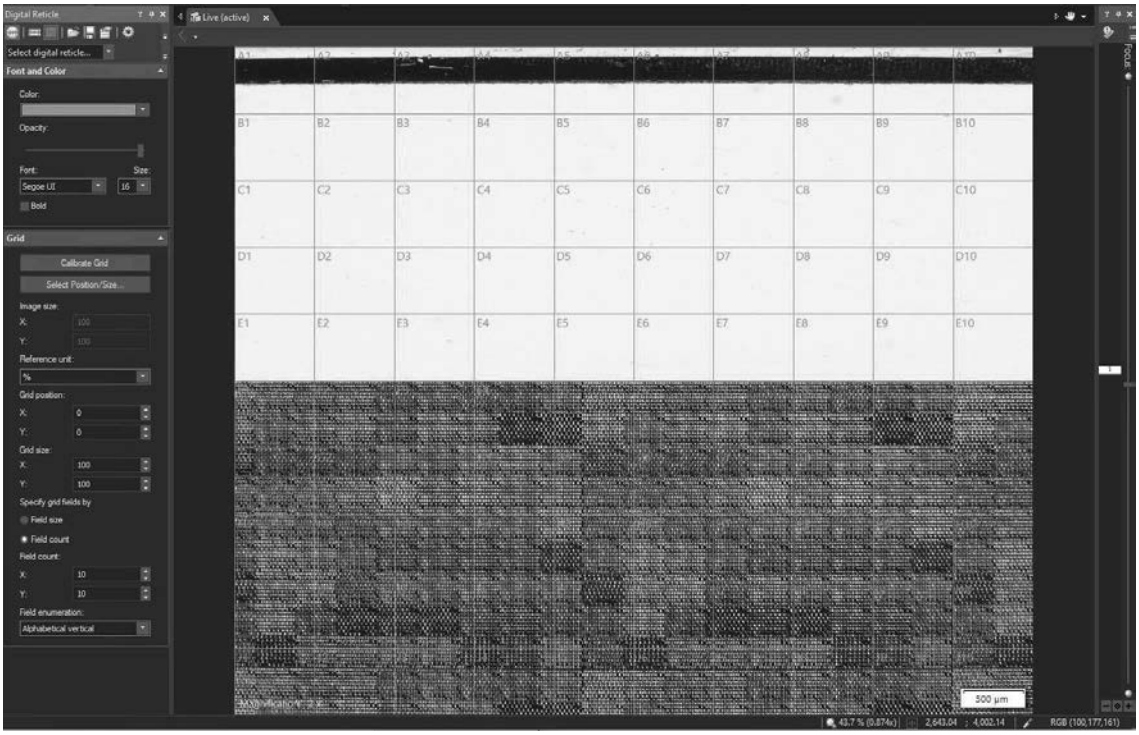
The units described below must be assembled and adjusted by Evident. If these units are assembled or adjusted by the customer, the operations are not ensured.

The color camera iDS (VS-264C) is adjusted ex works and locked on place with a locker. Therefore, a recalibration on site is probably not necessary. The following check of the color camera rotation must be performed by Evident before the initial start-up of the VS200 system.

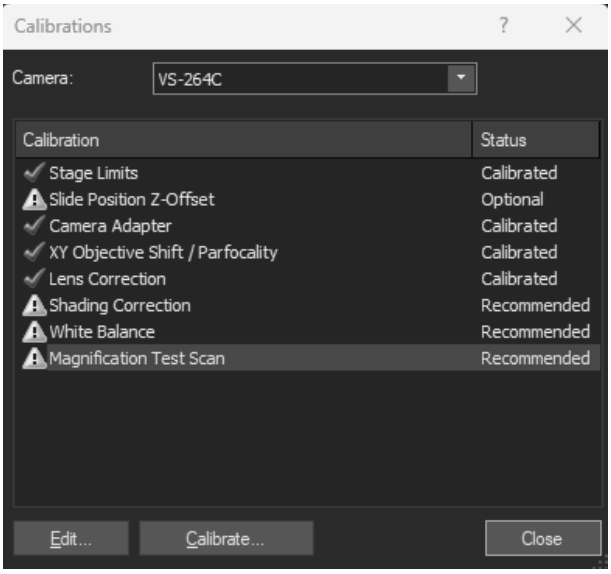
1. Load the calibration slide on the stage.
2. In the VS200 ASW software, go to the [Manual control] layout.
3. Use the [BF] observation method and start the live mode.
4. Activate the tool window [Digital Reticle]. To do so, click on [View] in the menu and select the [Digital Reticle] entry. You can change the format of the digital reticle and the color using the commands in the tool window.

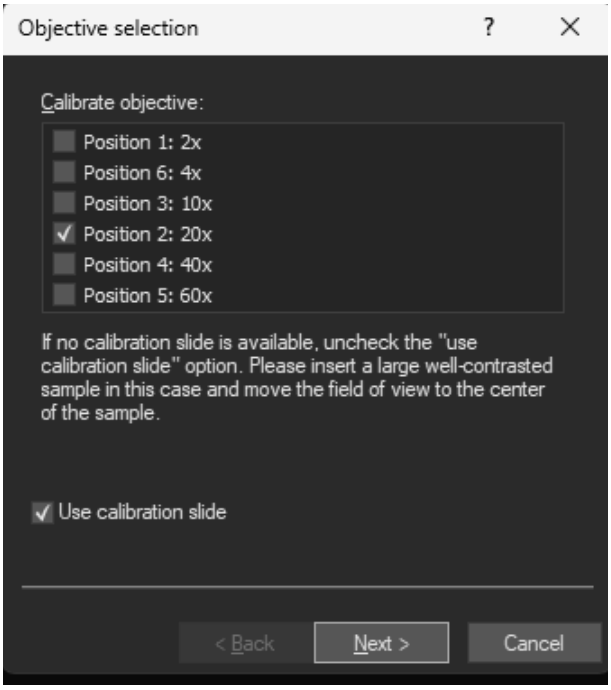


5. Navigate the slide in the Y direction, so the upper border of the letter matrix align to the central area of the field of view.
6. Loosen the headless hex screw that is used to fix the TV0.63 adapter at the flange of the beam splitter.
7. Rotate the TV0.63-adapter until the border of the letter matrix is perfectly parallel to one of the horizontal line of the digital reticle in the central area of the field of view. The letters in the letter matrix pattern must appear up-side down.



8. Tighten the fixation hex screw of the TV0.63 adapter and make sure that during this step the previously done alignment has not been corrupted. If this is the case, repeat steps 6 and 7.
9. Start the [Magnification Test Scan] calibration and run it only with the 20x objective.





10. Check the results of the [Magnification Test Scan] calibration. The rotation value must be in the range of +/- 0,05° (but it is recommended to reduce it as close to 0° as possible).

11. If the result is not yet satisfactory, repeat steps 6 to 10 until the rotation value is in the accepted range.

An adjustment of the color camera rotation requires a new calibration of all the other calibrations except the [Stage Limits] calibration and the [Slide Position Z-Offset] calibration for both the color and monochrome camera connected to the VS200 system. See Calibrate VS200 using the VS Calibration Slide on page 139 and Additional calibrations for a fluorescence system on page 186.

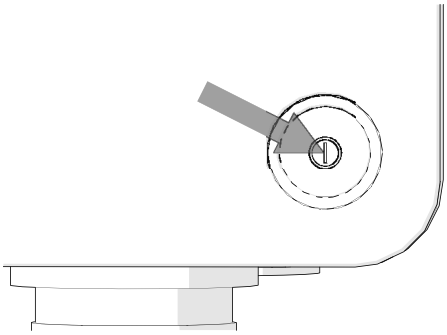
28.7 Adjusting the leveling feet of the VS200 loader

The units described below must be assembled and adjusted by Evident. If these units are assembled or adjusted by the customer, the operations are not ensured.

This chapter describes how to adjust the leveling feet of the VS200 loader. This adjustment is necessary when the VS200 scanner is operated with a VS200 loader and the height of both devices does not match. In this case, the VS200 scanner and the VS200 loader cannot be connected.

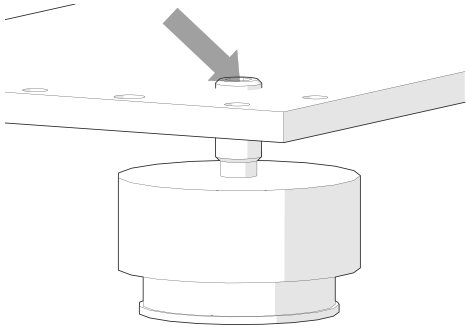
» Hex key (size 6 mm)

1. Switch the VS200 system off using the main power switch and disconnect the system from the power supply. To do so, disconnect the external power supply unit from the power supply.



2. Remove the housing rear cover.
3. To adjust the leveling feet, either turn the leveling feet directly or use a hex key (size 6 mm hex key). Insert the hex key as indicated in the figure.

   Adjust the leveling feet so that you can just slide the loader onto the connector's ground plate.



4. Mount the housing.
5. Reconnect the VS200 scanner to the power supply.

28.8 Barcodes

For further details on how to activate and use the barcode within the VS200 ASW software, please refer to the software's help system.

28.8.1 Supported barcodes

The VS200 system is able to detect barcodes on the label area of slides. The system supports 1D and 2D barcodes of the following types:

1D types	Codabar
	Code 39
	Code 128
	EAN-8
	EAN-13
	GS1 Databar
	GS1-128
	Interleaved 2 of 5
	UPC-A
	UPC-E
2D types	Data Matrix
	PDF-417
	QR Code

28.8.2 Quiet zones

Definition

The quiet zone is an area before and after the 1D barcode (left and right of it) which does not contain any text, graphics, markings, scratches, or other colors beside the barcode background color. For 2D barcodes, the quiet zone is both left and right of the barcode as well as above and below it. The quiet zones are necessary for a scanner to be able to clearly detect the beginning and the end of a barcode.

Correct sizes

Minimum size of quiet zones	Recommended sizes of quiet zones
10 times the size of the width of the smallest barcode module	15 times the size of the width of the smallest barcode module
2.5 mm	6.5 mm

Examples of correct sizes

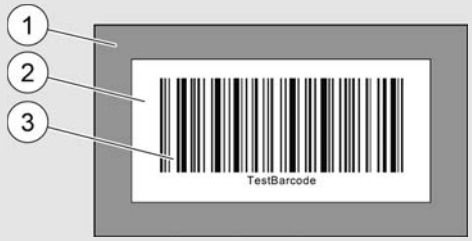
Example 1: The width of the smallest barcode module is 1 mm. The quiet zone shall be at least 10 mm.

Example 2: The width of the smallest barcode module is 0.1 mm. The quiet zone shall be at least 2.5 mm.

Correct positioning of the barcode on the slide

To ensure that the barcode capture functions correctly, the complete barcode and the full quiet zone around the barcode have to be well positioned on the label area of the slide so that they are easy to capture by the device. If the barcode label is positioned too far to one side, the quiet zone is truncated and the barcode cannot be read by the device. The barcode also cannot be read out by the device if there is any additional writing, print, or other markings on the labels as these also interfere with the area of the barcode and the quiet zones.

Example



1 label area
There is no additional writing, print or other markings.

2 quiet zones
The quiet zones are the correct sizes.

3 barcode
The barcode is well positioned. There is no additional writing, print or other markings.

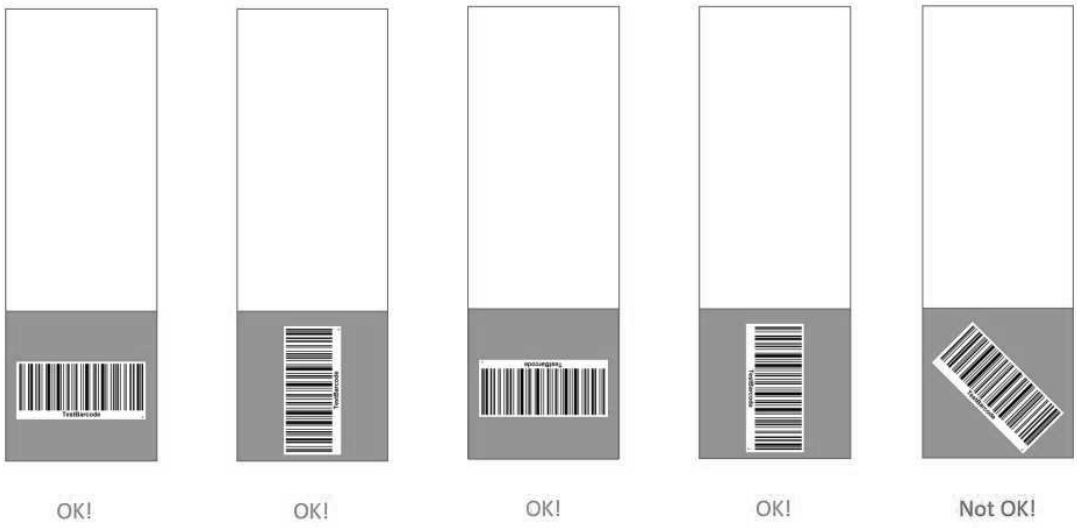
28.8.3 Barcode quality and orientation

It is recommended to print the barcode with a professional label printer of sufficient resolution. A minimum resolution of 300 DPI is strongly recommended.

In any case, the user must choose appropriate label printing settings and label paper to ensure a sufficient barcode quality.

When putting the barcode on the slide, it must not be rotated / tilted too much with respect to the slide edges.

The VS200 system supports all orthogonal barcode directions (with a tolerance of +/- 12 degrees). The system might not be able to correctly detect the barcode if the rotation is larger. See the following figure.

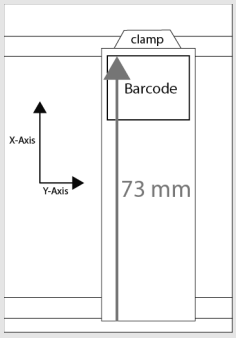


28.8.4 Restrictions

Using multiple barcodes on the same label is not supported by the system. If the system detects multiple barcodes in the configured label area, it will only keep the barcode with the highest, automatically determined confidence. The system will output a warning message (in batch scans, this message is written to the log file as well).

Not OK!

To protect your system's optical hardware, a small area next to the clamp that fixes the slide can't be scanned. Normally there is a label or a barcode next to this area. Make sure that there is sufficient space between the label or barcode and the clamp. The far edge of the label (or barcode) should not be more than 73 mm from the opposite edge of the slide. This ensures that it can be scanned in its entirety. Otherwise a 2D barcode would not be scanned in its entirety and could then not be read out successfully.



The example shows a 1x3 inch slide that has been placed in the tray. The red arrow shows a distance of 73 mm. This is the maximum distance that the far edge of the barcode or label is permitted to be from the opposite edge of the slide.

29 Preparing for transportation

Switch the VS200 system off using the main power switch and disconnect the system from the power supply. To do so, disconnect the external power supply unit from the power supply.

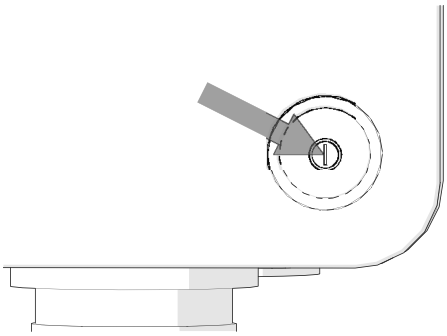
29.1 General preparation

1. Remove all trays from the base unit and loader.
2. Remove all objectives.
3. Remove the liquid light guide.
4. Remove the monochrome camera.
5. Pack all items securely.

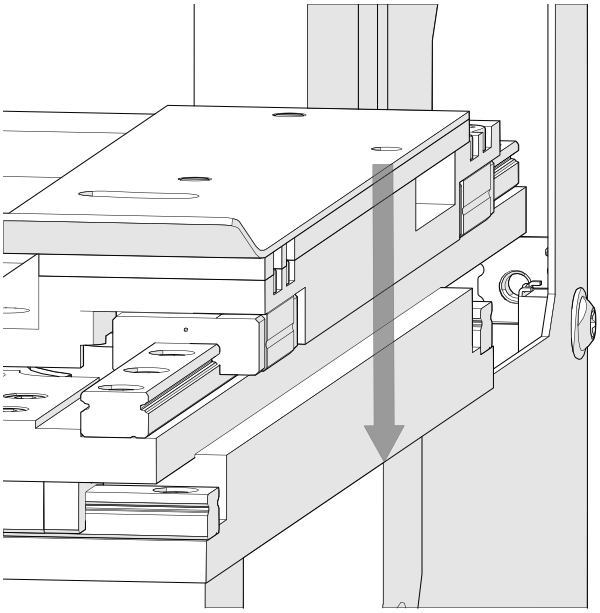
29.2 Mounting the transportation lock on the stage

The units described below must be assembled and adjusted by Evident. If these units are assembled or adjusted by the customer, the operations are not ensured.

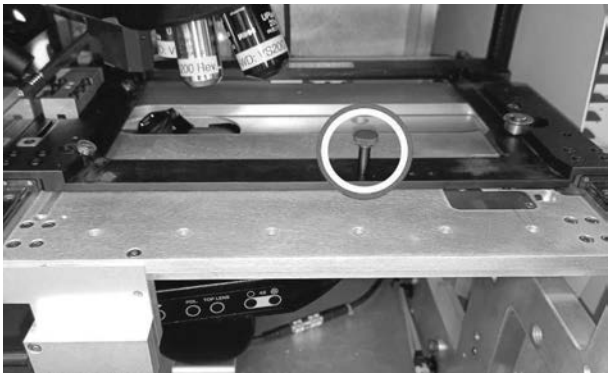
1. Switch the VS200 system off using the main power switch and disconnect the system from the power supply. To do so, disconnect the external power supply unit from the power supply.



2. Remove the following elements of the housing. You can remove more if required. You can find more information in the VS200 Repair Manual.
   » Right housing side cover
3. If necessary, manually move the upper moving plate of the stage so far to the right that all parts of the stage are right-aligned. To move the stage, grasp the top of the stage where the trays are inserted.



4. Slide the black frame of the stage forwards until the locking screw can be inserted (see figure below). Secure the stage by hand-tightening the locking screw shown.

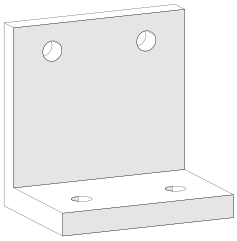


5. Mount the housing.

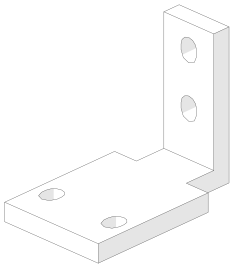
29.3 Mounting the transportation lock on the VS200 loader's tray hotel

The units described below must be assembled and adjusted by Evident. If these units are assembled or adjusted by the customer, the operations are not ensured.

This chapter describes how to mount the two transportation locks that secure the tray hotel in the VS200 loader.



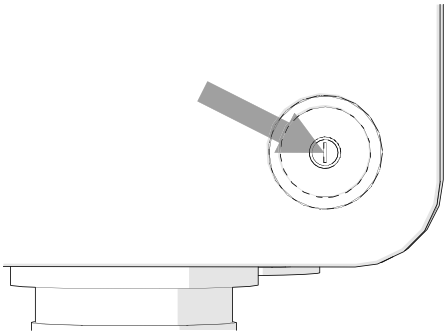
Transportation lock (1)



Transportation lock (2)

» Hex screwdriver (size 3 mm)

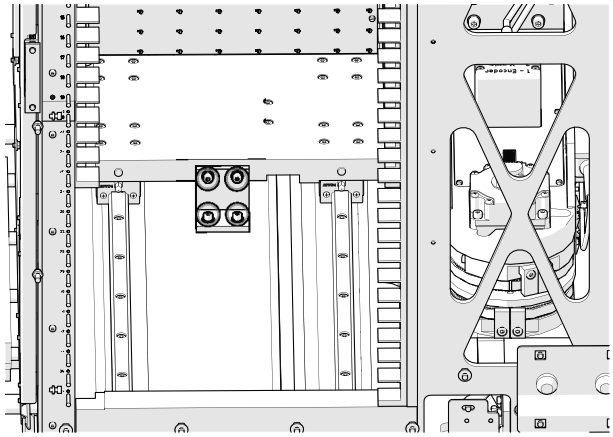
1. Switch the VS200 system off using the main power switch and disconnect the system from the power supply. To do so, disconnect the external power supply unit from the power supply.



2. Remove the housing.
3. Move the VS200 tray hotel into the park position all the way at the back. To do so, grasp the bottom plate of the VS200 tray hotel.

> Make sure that the gripper that is attached to the SCARA robot arm doesn't collide with the tray hotel.

4. Attach the transportation locks to the tray hotel. To position the transportation locks, refer to the following figures. Use the 4 hex screws (size 3 mm hex screwdriver) to attach the lower transportation lock to the tray hotel as well as to the bottom panel of the VS200 loader.



5. Now attach the upper transportation lock that connects the top right of the tray hotel with the right of the frame. To do so, use the 4 hex screws (size 3 mm hex screwdriver) indicated in the figure. Make sure that the surfaces of the bracket are flush with the vertical frame structure and the horizontal top of the tray hotel before tightening the screws. Tighten the screws that are screwed into the vertical frame structure first and the other two screws last.