User's Manual

CellVoyager CV7000

High-throughput Cytological Discovery System

IM 80H01C01-01E

YOKOGAWA

Yokogawa Electric Corporation

IM 80H01C01-01E
14th Edition

Introduction

Thank you for purchasing the CellVoyager CV7000.
This user's manual describes the functions, operating procedures, and safety and handling precautions of the CV7000. Before you start to use the CV7000, please read this manual carefully to enable correct use of the system. For more information on the specifications, refer to the technical specifications that are published separately.
After reading this manual, keep it in a handy place so that you can refer to it whenever necessary.

Notes

- Unauthorized reproduction or reprinting of this manual in whole or in part is prohibited.
- The information contained in this manual is subject to change without prior notice due to improvements in performance and functionality or for other reasons.
- This manual has been prepared with the utmost care; however, if you have any questions, or note any errors, please contact us.
- Follow the operating instructions to ensure that the system remains stable.

Trademarks

- CellVoyager and CSU are registered trademarks of Yokogawa Electric Corporation.
- Microsoft and Windows are registered trademarks or trademarks of Microsoft Corporation in the United States and other countries.
- The CellVoyager is using patents on High Content Screening and High Content Analysis of Thermo Fisher Scientific Inc.
- In this document, registered trademarks or trademarks are not indicated by™ or ®.

Relevant Documents

- IM 80H01C03-01E CV7000 Analysis Software User's Manual
- IM 80H01A16-01E Image Correction Software User's Manual
- IM 80H01A17-01E Postprocessor User's Manual

IM 80H01C01-01E 14th Edition: August 2016(YK)

History

June 2012 1st edition issued.

September 2012 2nd edition issued.

Modification
Added the explanation about camera gain setting.

November 2012 3rd edition issued.

Modification
Added camera binning 2 × 2 for sCMOS.
Added setting items for source plate name and source plate information in the “Start Measurement” screen when dispensing measurement.
Added the setting items for postprocessor function.

February 2013 4th edition issued.

Modification
Added the explanation about plate information.

October 2013 5th edition issued.

Modification
Added the AirBlow function in the dispensing setting file. Added Plate Bottom for tip position.
Added the SUM function for Image Processing in the 3D Fluorescence Acquisition and Z-stack Bright-field/Phase-contrast Acquisition screens.
Added snapshot function in the measurement result sub screen.
Added search filter function in the Measurement Setting List screen.
Modified the screen for high-speed time-lapse setting.

November 2013 6th edition issued.

Modification
Added the explanation about QUAD-DM model.
Modified bright-field acquisition to match pixel position with each image of confocal or epifluorescent imaging.
Added the explanation for 384-tip rack model.

December 2013 7th edition issued.

Modification
Added the function to record CO2 concentration and temperature logs.
Added XY offsets in the “Add Acquisition Points” screen.

June 2014 8th edition issued.

Modification
Added the function to perform high-speed time-lapse imaging with multi dispensing.
Added the function to expansion the Acquisition Point Screen. Changed the operability of field selection.
Added the explanation of slide glass holder.

November 2014 9th edition issued

Modification
Added binning 3x3 and 4x4.
Added the function to perform high-speed time-lapse Imaging working with normal Imaging.

January 2015 10th edition issued

Modification
Added procedure for registering and selecting fluorophore.
Added explanation of high precision autofocus mode.
Added explanation of performing autofocus during high-speed time-lapse imaging.
Modified the explanation of slide glass holder.

April 2015 11th edition issued

Modification
Added “Relevant Documents”
Added explanations about partial tiling function.
Deleted explanations about Camera Setting (Camera gain).
Added cautions of sample setting
Added explanation about changes of CO_2 concentration control

October 2015 12th edition issued

Modification
Added explanations about time line copying function.
Modified explanations about CO_2 concentration setting of stage incubator.
Added explanations about imaging forbidden area of using water immersion objective lens.
Modified explanations about slide glass measurement using water immersion objective lens.
Added explanations about display of number of times that dispenser syringe moves.
Modified explanations about saving path of measurement files.

June 2016 13th edition issued

Modification
Added explanations about digital phase contrast (DPC) acquisition function.
Modified explanation about Stage Incubator setting.
Added Well plate registration function.
Added explanation about On-the-fly image correction function.

August 2016 14th edition issued

Modification
Modified description for WEEE directive.
Added table of MS code

For Safe Use of This Equipment

To ensure safe and correct use of this product, be sure to follow the precautions below. If this product is used in a manner not specified by this manual, the protection provided by this product may be impaired. If you do not follow the precautions when handling the product, we do not guarantee the safety and product.

The following safety symbols are used for this product.


WARNING or 
CAUTION

These symbols indicate that you need to handle the equipment with care. These symbols are displayed in locations where you need to refer to the user's manual to protect your safety and the safety of equipment.

These symbols in the manual are used in the following cases.


PROTECTIVE GROUND TERMINAL

This symbol indicates a protective ground terminal. Be sure to provide a ground connection before turning on the equipment to prevent an electric shock.


ALTERNATING CURRENT

This symbol indicates an alternating current.

POWER ON

This symbol indicates POWER ON.


POWER OFF

This symbol indicates POWER OFF.


WARNING

On the equipment, this symbol is displayed in the locations where you must refer to the manual to protect yourself and the equipment from serious accidents. In the manual, this symbol is placed near text that describes the precautions to help avoid hazardous situations that could result in a bodily injury or death of the user, such as an electric shock, or that could result in damage to the equipment and devices.

⚠ CAUTION

On the equipment, this symbol is displayed in the locations where you must refer to the manual to protect yourself and the equipment from accidents. In the manual, this symbol is placed near text that describes the precautions to help avoid hazardous situations that could result in a minor bodily injury of the user, or that could result in damage to the equipment.

⚠ WARNING

- Be sure to turn on the power to the equipment after confirming that the power-supply voltage of the equipment matches the voltage of the supplied power.
- This is a Class 1 laser product. However, the equipment houses a Class 3B laser, which is protected by the enclosure and the interlocks provided at openings. When using this product, heed the precautions explained in “Laser Products Handling Precautions.”
- Inhalation of 5%CO_2 gas may be harmful to the human body. Be sure to provide ventilation, lead exhaust gas from the incubator to outside of the room, or take other appropriate measures.
- Do not use the equipment by wet hand. It may cause electric shock and suffer electrical or mechanical damages.
- Do not bring electrically charged objects near the signal terminals. Equipment failure may result.
- Do not pour volatile chemicals onto the display or keep it in contact with rubber or plastic products for an extended period of time.
- Do not give shock to this equipment.
- Do not block the vent openings.
- Should you notice smoke coming out of the main unit, foul smell, abnormal noise or any other abnormality, immediately turn off the power switch and cut off the power supply. Contact the Yokogawa dealer from which you have purchased the equipment or Yokogawa's service department to notify the abnormality.
- To stop the equipment immediately in case of emergency, press the EMERGENCY STOP button. To release the button, turn it clockwise.

⚠ CAUTION

- Use of controls or adjustments or performance of procedures other than those specified herein may result in hazardous exposure.
- Use the I/O terminals of the equipment within the range of the specifications to prevent damage to the equipment.
- This product uses precision optics. Therefore, do not use the equipment in locations where there are large vibrations, a lot of dust, high humidity and high temperature (in places near heating equipment or exposed to direct sunlight), rapid changes in the temperature (dew condensation), or corrosive or combustible gases.
- Never touch any of the internal parts of this product. The optical system housed in the product may become dirty, damaged or out of calibration, etc., leading to equipment failure.
- If the equipment malfunctions, contact us without attempting to access the inside or take any other action to resolve the problem yourself.

WEEE (Waste Electrical and Electronic Equipment), Directive

(This directive is only valid in the EU.)

- This instrument complies with the WEEE Directive marking requirement. The marking above indicates that you must not discard this instrument in domestic household waste.
- Product Category
  With reference to the instrument type in the WEEE directive Annex 1, this product is classified as a “Monitoring and Control instrument” product.
- When disposing this instrument in the EU, contact the distributer whom you bought it from. Do not dispose in domestic household waste.

Warning and Caution Labels

The safety warning and caution labels attached on this product are listed below.

Warning Label (1)

Warning Label (2)

Warning Label (3)

Warning Label (4)

Caution Label (1)

Caution Label (2)

Caution Label (3)

Warning Label (1)
This label warns you to be careful because there is a risk that your hand or clothes may get caught. This label warns you not to approach the spot during operation.

Warning Label (2)
This label indicates a risk that your hand may get caught. This label warns you not to insert your hand.

Warning Label (3)
This label indicates a risk of an electric shock and warns you not to touch the conductor.

Warning Label (4)
This label indicates a risk of laser radiation. Do not stare into beam.

Caution Label (1)
This label warns that safety management is required to use the laser. This label indicates a risk of your eyes or skin being exposed to a class 3R laser beam if you open this part.

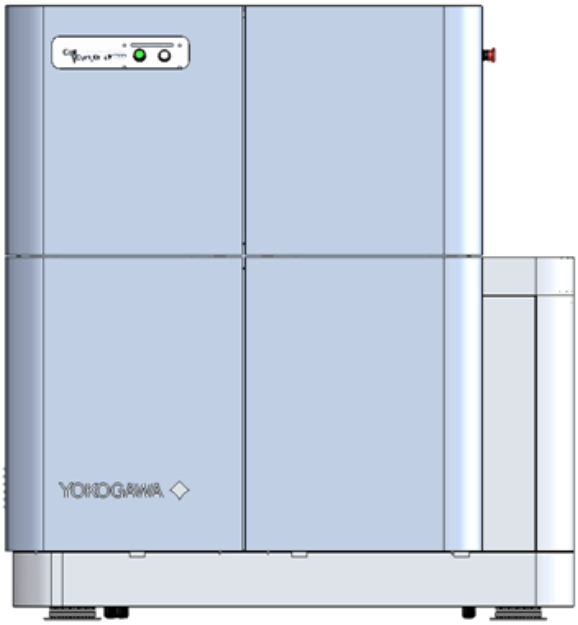
Caution Label (2)
This label warns that safety management is required to use the laser. This label indicates a risk of your eyes or skin being exposed to a class 3B laser beam if you open this part.

Caution Label (3)
This label indicates that it is necessary to refer to this manual.

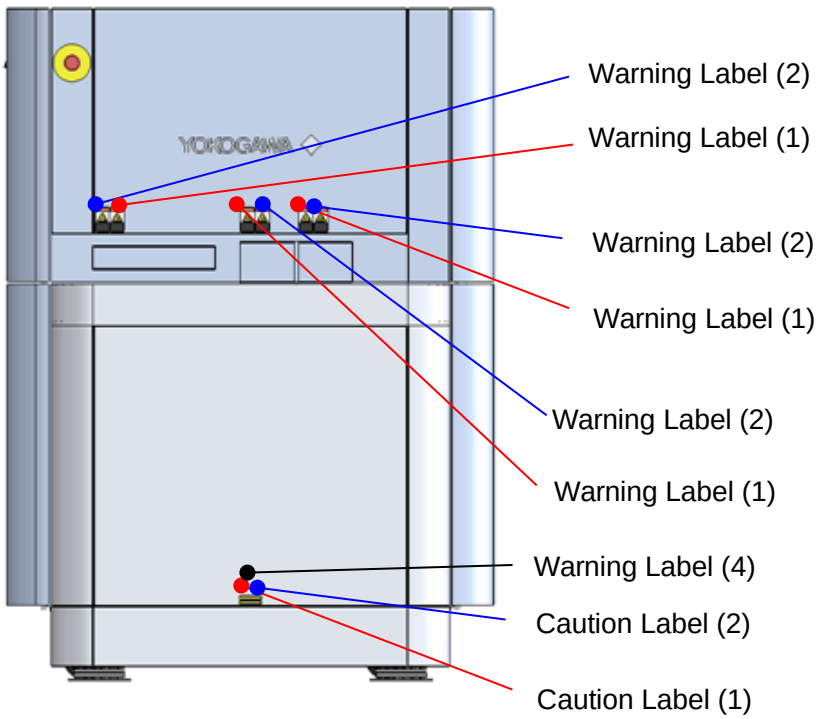
Locations of Warning Labels

Warning labels are placed in locations where safety management is required to use this product. The following shows the placed warning labels and their placement positions. If the equipment malfunctions, contact us without attempting to access the inside or take any other action to resolve the problem yourself.

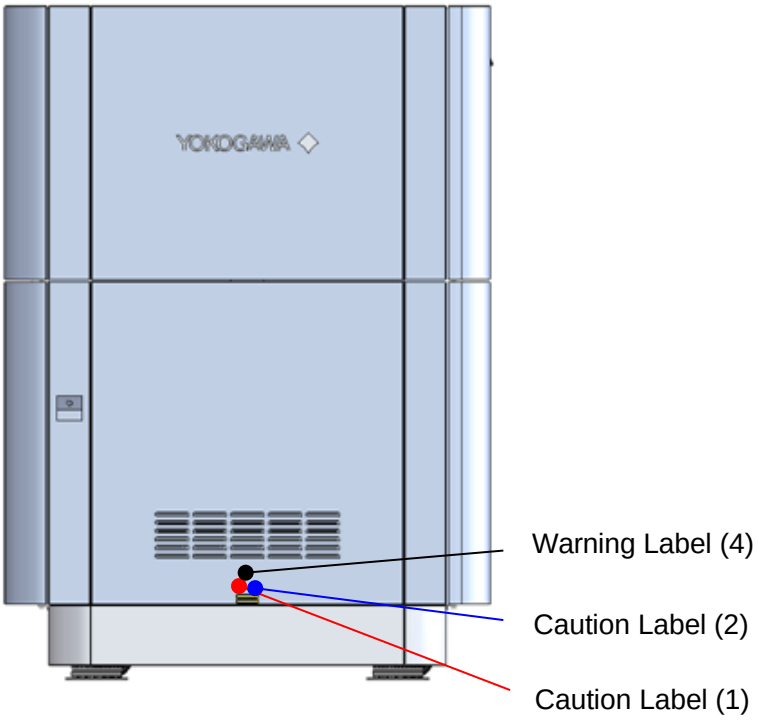
Front Panel



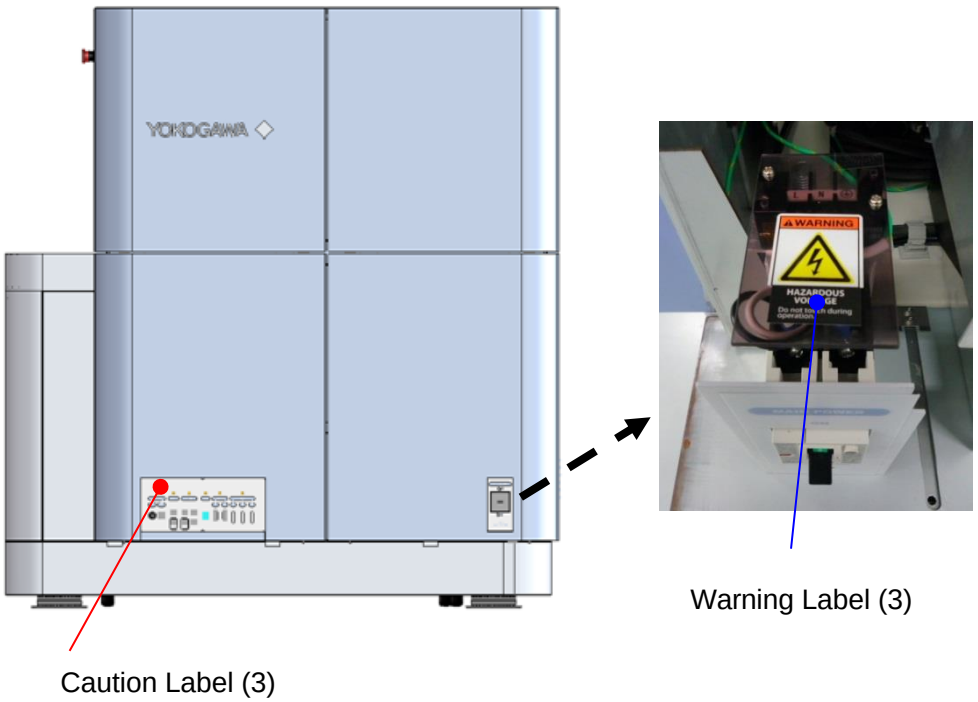
Right Side Panel



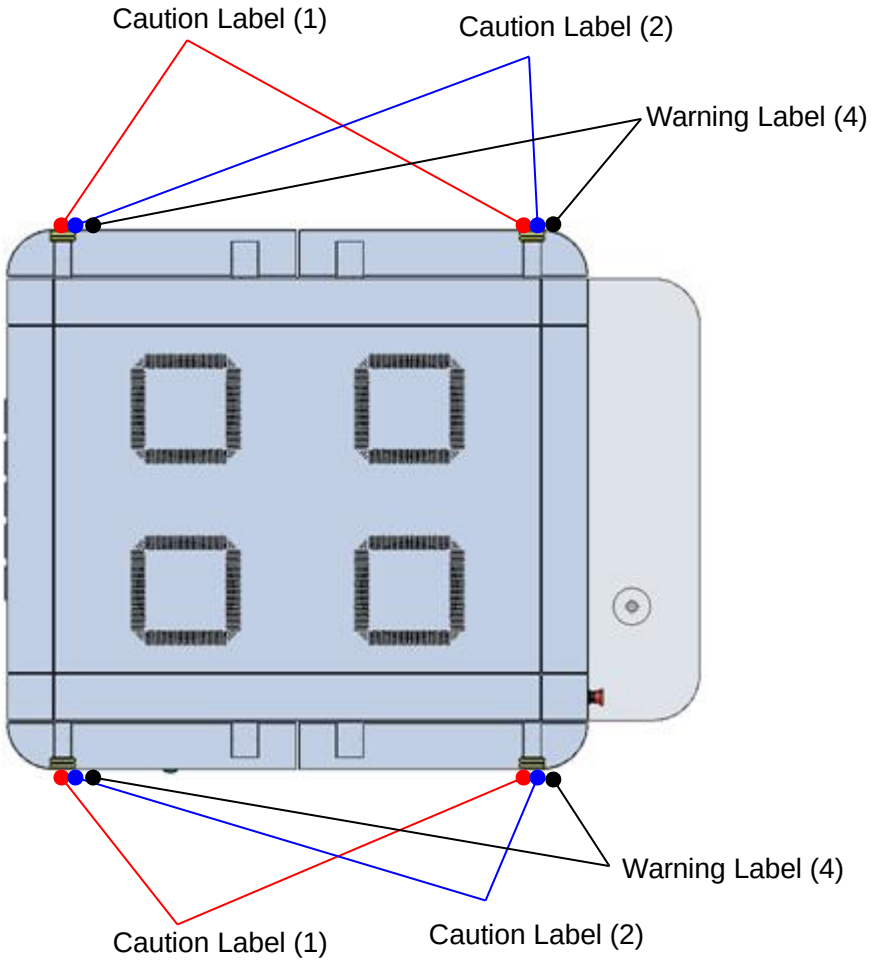
Left Side Panel



Rear Panel



Top Panel


Caution Label (1)
Caution Label (2)
Warning Label (4)
Warning Label (4)
Caution Label (1)
Caution Label (2)

Laser Product Handling Precautions

Laser products are classified based on their exposure emission limit determined by the wavelength and power characteristics of laser beam. A different set of common safety standards applies to each class of products. The product explained herein belongs to Class 1. However, the equipment houses a Class 3B laser, which is protected by the enclosure and the interlocks provided at openings.

Only service personnel can remove the covers on which a warning label is attached. If these covers are removed, class 3B laser beam will be emitted. Directly looking into Class 3B laser beam or beam reflected on a mirror may cause eye damage and is extremely dangerous. Exercise due caution when handling this product.

Safety Standards

A class 1 laser is safe under all conditions of normal use. This means the maximum permissible exposure cannot be exceeded. This equipment is designed in accordance with the IEC60825-1 Radiation safety standards for laser products and these lasers must be labeled with the following label, but are exempt from the requirements of the Laser Safety Program.

CLASS 1 LASER PRODUCT
IEC60825-1 : 2007

Indemnity

- Yokogawa shall provide no warranty regarding this product, unless otherwise specified separately in “Warranty Terms.”
- Yokogawa shall assume no responsibility for any loss suffered by a customer or third party as a result of use of this product, or any loss or indirect loss suffered by a customer or third party due to a defect in this product or any other problem not predictable by Yokogawa.

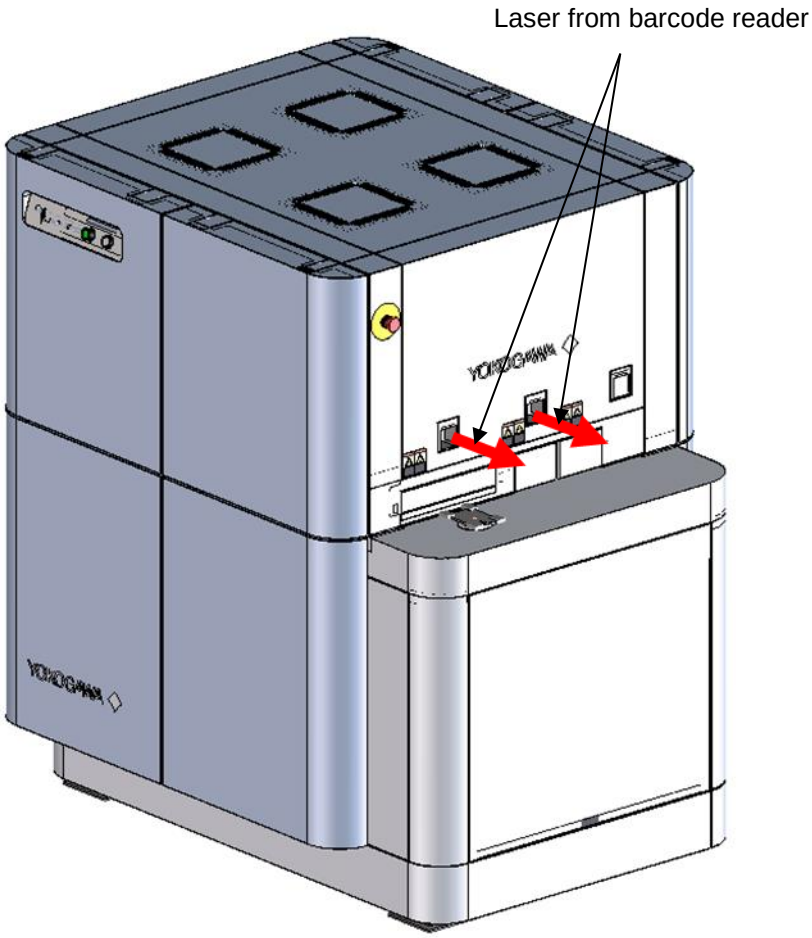
Laser Specification

Barcode Reader

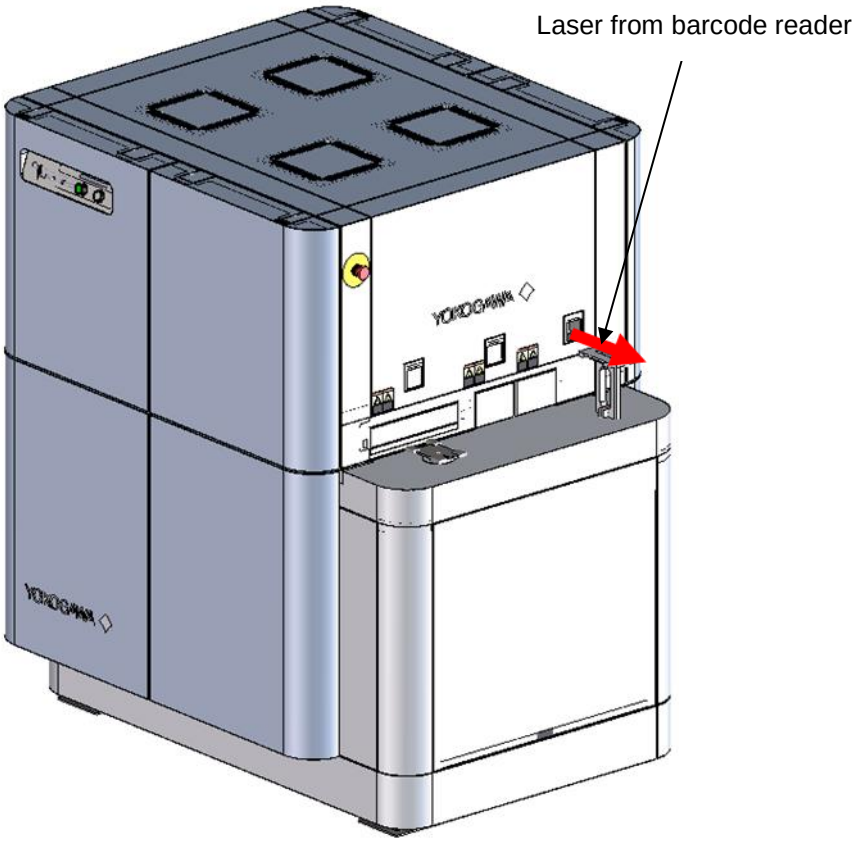
This product emits the laser below if it has barcode option.

Wavelength: 660 nm
Output: 0.09 mW
Pulse width: 200 μs
Beam divergence: parallel beam
This laser beam is not for barcode reading.
This laser beam is only for aiming beam and is only used at installation time.

(1) 2-barcode reader option



(2) 1-barcode reader option



Microscope

This product has lasers below in it.

(1) 405 nm laser
Wavelength: 405 ±5 nm
Output: 100 mW
Beam divergence: 0.4 mrad
Laser type: Continuous

(2) 488 nm laser
Wavelength: 488 ±2 nm
Output: 200 mW
Beam divergence: 1.2 mrad
Laser type: Continuous

(3) 532 nm laser
Wavelength: 532 ±2 nm
Output: 200 mW
Beam divergence: 1.3 mrad
Laser type: Continuous

(4) 561 nm laser
Wavelength: 561 ±2 nm
Output: 200 mW
Beam divergence: 1.3 mrad
Laser type: Continuous

(5) 640 nm laser
Wavelength: 640 +4/-5 nm
Output: 100 mW
Beam divergence: 1 mrad
Laser type: Continuous

(6) 785 nm laser
Wavelength: 785 +15/-10 nm
Output: 2.5mW
Beam divergence: parallel beam
Laser type: Continuous

Applicable Standards

CE Marking

●EMC Directives:

EN 61326-1 Class A, Table 1 (Basic immunity requirements)

Electrical equipment for measurement, control and laboratory use - EMC requirements -

Part 1: General requirements

●Machinery Directive:

ISO12100

Safety of machinery

- General principles for design - Risk assessment and risk reduction

EN 13849-1

Safety of machinery

- Safety-related parts of control systems - Part 1: General principles for design

EN 61010-1

Safety requirements for electrical equipment for measurement, control, and laboratory use, Part 1: General requirements

EN 60825-1

Safety of laser products, Part 1: Equipment classification and requirements

EC DECLARATION OF CONFORMITY

YOKOGAWA For CE

High-throughput Cytological Discovery System
Model: CV7000

Manufactured by

Yokogawa Electric Corporation
2-9-32 Nakacho,
Musashino-shi,
Tokyo, 180-8750
Japan

Means of Conformity

The Product is in conformity with EC law as approximated by the Machinery Directive 2006/42/EC, based on Technical Documentation File No.**2193 Issue 1, Revision 0**, November 2013

Standards used as guidance
Machinery Directive

EN ISO 12100: 2010
EN ISO 13849-1: 2008
EN 60825-1: 2007
EN 61010-1: 2010
Refer to complete listing in Technical File

The Machinery Directive Technical File compiled from manufacturers documentation and held in the EU, on behalf of the manufacturer by

TRaC Global Ltd
100 Frobisher Business Park,
Leigh Sinton Road, Malvern,
Worcestershire, WR14 1BX
United Kingdom

Signature of Responsible Person:

Takayuki Kei

Mr. Takayuki Kei
Technical General Manager
Life Science Headquarters R&D Department
Yokogawa Electric Corporation

Date: **21st November 2013**

Document No. **TRA-015425-00 DofC Issue 1**

Doc. No.: EEN302-C03

EC DECLARATION OF CONFORMITY

We **Yokogawa Electric Corporation**
2-9-32 Nakacho, Musashino-shi, Tokyo, 180-8750 Japan

declare under our sole responsibility that the product

CV7000 **High-throughput Cytological Discovery System**
(See Appendix for the detailed type designation)

to which this declaration relates is in conformity with the following standards or other normative documents:

EN 61326-1: 2013 Class A, Table 1 (Basic immunity requirements)
Electrical equipment for measurement, control and laboratory use – EMC requirements-
Part 1: General requirements

following the provisions of EMC directive 2004/108/EC.

Subject products are manufactured and tested according to appropriate quality control procedures.

Tokyo, 31 July, 2015

Signature:

Takayuki Kei

Takayuki Kei
R&D Section Manager
Life Science Headquarters
Yokogawa Electric Corporation

Yokogawa Electric Corporation

How to Use This Manual

This user manual consists of Chapters 1 to 14, the details of which are explained below.

Chapter	Title and content
1	Overview of the Equipment A functional overview of this equipment is explained.
2	Before Use Installation and wiring methods are explained.
3	Starting and Shutting Down the Equipment The name of each part and starting/shutdown of the equipment are explained.
4	Entering Well Plate Information Files Entry of well plate information is explained.
5	Using the Measurement Software Functions and operations of the measurement software are explained.
6	Explanation of Measurement Software Screens Screens of the measurement software are explained.
7	Setting Examples of Measurement Setting Files Examples of measurement using this equipment are explained.
8	Measurement Measurement operations are explained.
9	Checking Measured Images Checking of captured images is explained.
10	Saving Measured Files Where to save captured images is explained.
11	Troubleshooting Troubleshooting methods for this equipment are explained.
12	Maintenance and Inspection Maintenance and inspection of this equipment are explained.
13	Warranty The terms of warranty applicable to this equipment are explained.
14	General Specification The general specification about CV7000 is explained.

Content

1. Overview of the Equipment

1.1. About the CV7000

In basic research fields of medicine, biology, pharmacology, agriculture, etc., as well as applied research fields such as drug discovery, advancement of bio-research is making “live cell imaging” a general approach to observing live cells for a long period.
This equipment is a high-throughput cytological discovery system designed to let the users study various reactions of live cells both quickly and in detail to increase the efficiencies of drug development, compound evaluation, cell function study, etc.

Features of the CV7000

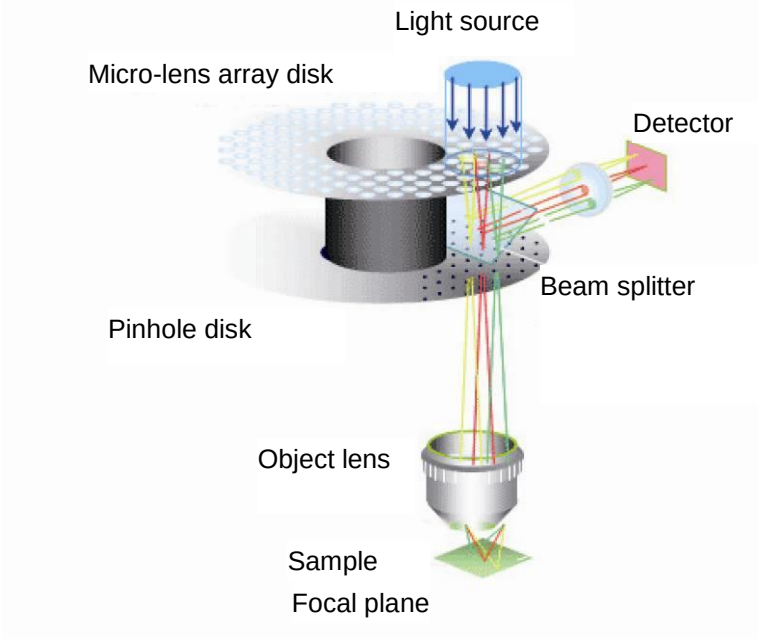
1. The industry’s most advanced live cell observation function
   The CV7000 is equipped with a confocal scanner unit CSU that uses the multi-scan system to minimize cell damage due to laser irradiation.

2. The industry’s fastest screening
   High-speed precision positioning, high-speed auto-focus and high-speed image acquisition technologies are combined to achieve high throughput.
   High-speed image acquisition is attained at less than 60 sec. for a 96 wellplate.

3. One of the industry’s best optical technologies
   The CV7000 adopts the high-resolution real confocal system using Nipkow disks with micro-lens array. The user can also freely change the observation mode among confocal, epifluorescence and phase contrast.
   Fourfold wide-field images can be acquired by using the confocal scanner unit designed for wide-field imaging. (Compared to the CV6000)

1.2. Operating Principle of the Nipkow Disk System with Micro-lens Array

Two disks, including the “pinhole array disk” having many pinholes arranged in a helical pattern, and the “micro-lens array disk” that condenses excited laser to individual pinholes, are operated jointly at high speed to perform multiple scans over the observation area with approximately 1,000 laser beams.
Multiple beam scans are performed not only at high speed, but also with each beam exciting fluorochromes at high efficiency and very low laser intensity. This results in an optimal live cell observation system where phototoxicity and fluorescence photobleaching are suppressed notably compared to any conventional system.



Operating Principle of the Nipkow Disk System with Micro-lens Array

2. Before Use This Equipment

2.1. Installing This Equipment

Installation Conditions

The following utilities are required.

Main unit

Power supply : 230VAC 50Hz 2kVA max
Grounding resistance : 100 ohm or less
CO_2 : Purity 99.95% or higher 0.2L/min or more 0.3MPa
Air : Cleanliness grade, Humidity and moisture content grade 6(dew point underpressure 10℃ or below), Solid grain grade 2, oil grade 2, foul smell removed by active carbon filters, 5L/min or more 0.6MPa

Workstation

Power supply : 100V-240VAC±10% 50/60Hz 1.5kVA max

Installation Location

Install this equipment in a location meeting the conditions specified below.

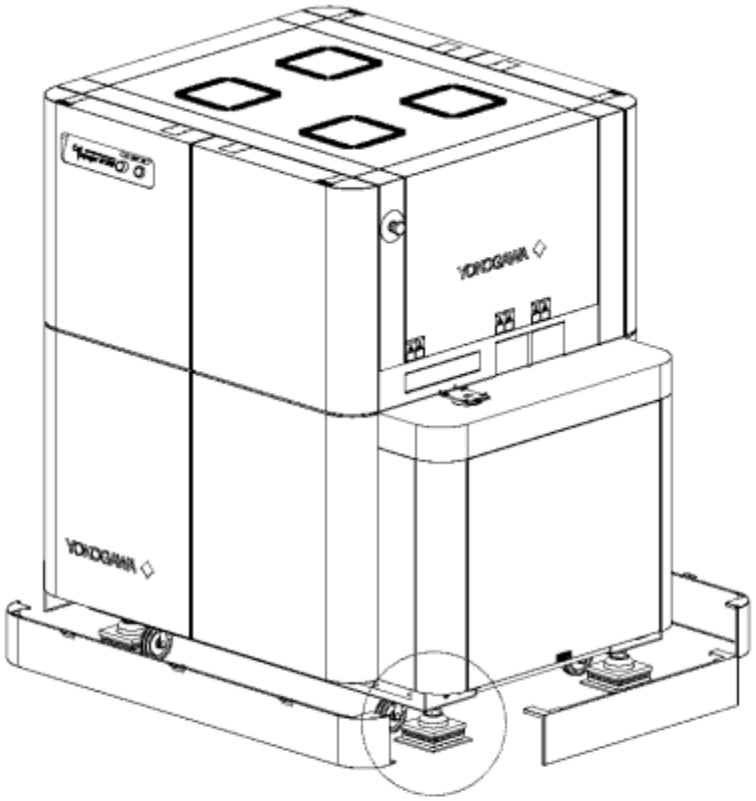
- Location where enough space is available
  Installation of this equipment requires a space of at least 2900 W x 3000 D x 2550 H (mm).
- Location subject to minimal mechanical vibration
  Install this equipment by selecting a location subject to minimal mechanical vibration.
- Level location
  When installing this equipment, make sure the equipment does not tilt to the left or right, but remains level.
- Location where the floor has sufficient strength
  This equipment weighs 650 kg. Install it in a location where the floor is strong enough to withstand this load.
- Location subject to minimal lamp soot, steam, dust, corrosive gases, etc.
- Install this equipment in a location where height above sea level is less than 1000m.

Note
Moving the equipment from a hot, humid location to a high-altitude location or otherwise subjecting the equipment to a sudden temperature shift may cause bedewing.

Installation Environment

Ambient operating temperature range : 15 to 30℃
Ambient operating humidity range : 10 to 70%RH non-condensing

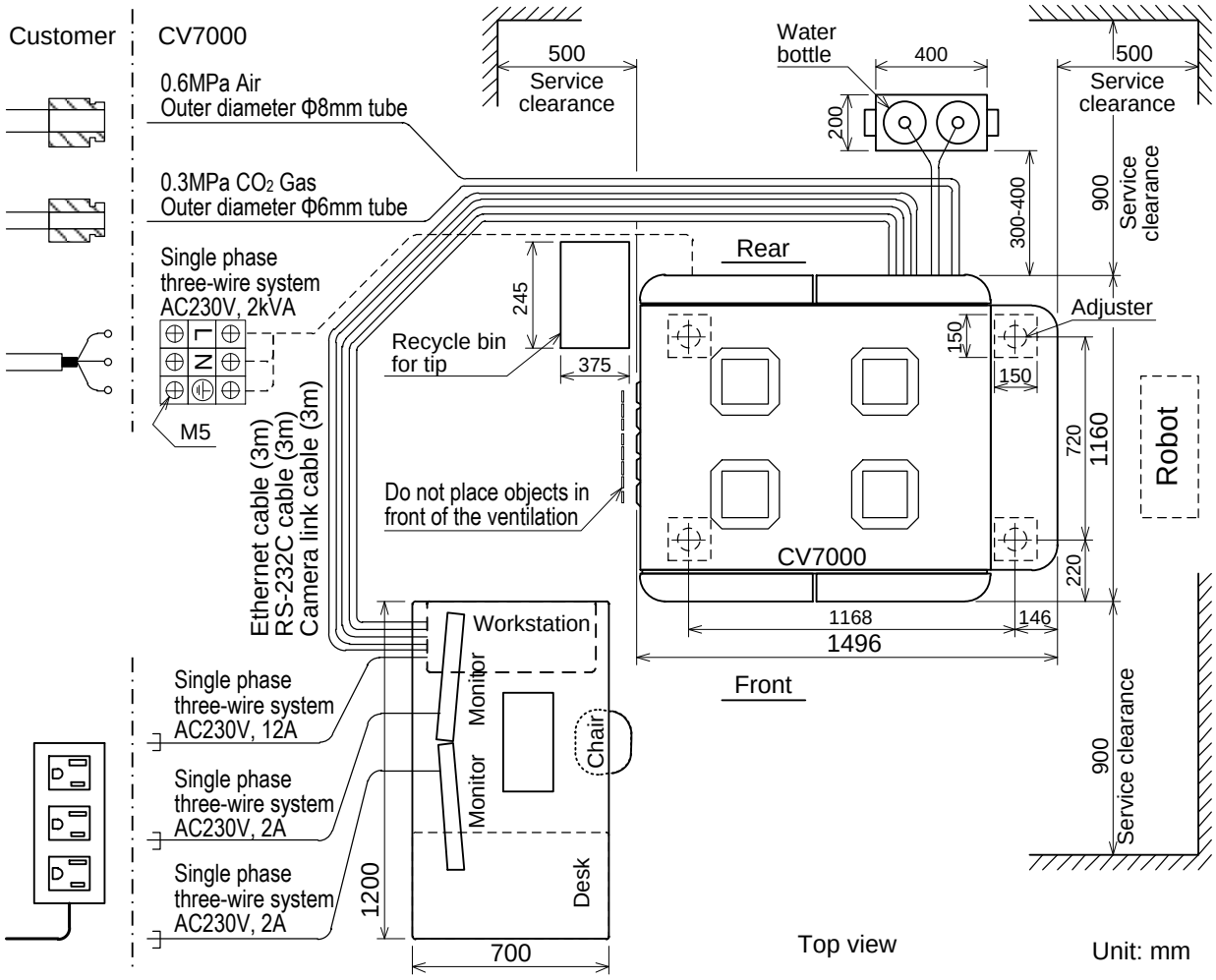
Main Unit installation



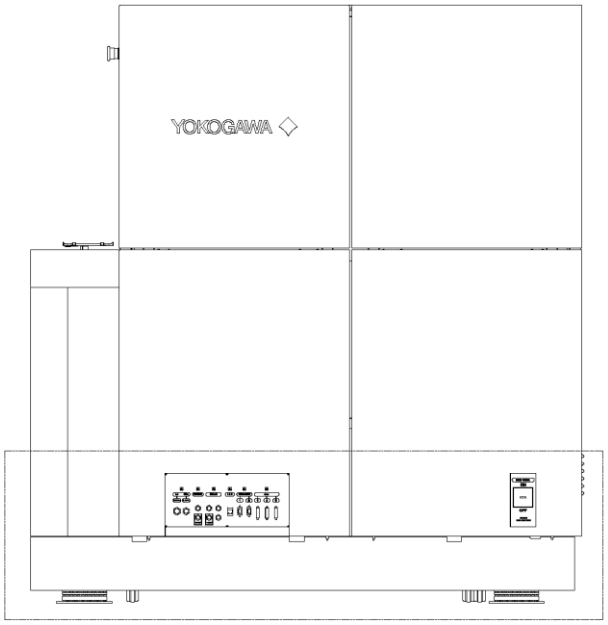
2.2. Connecting the System

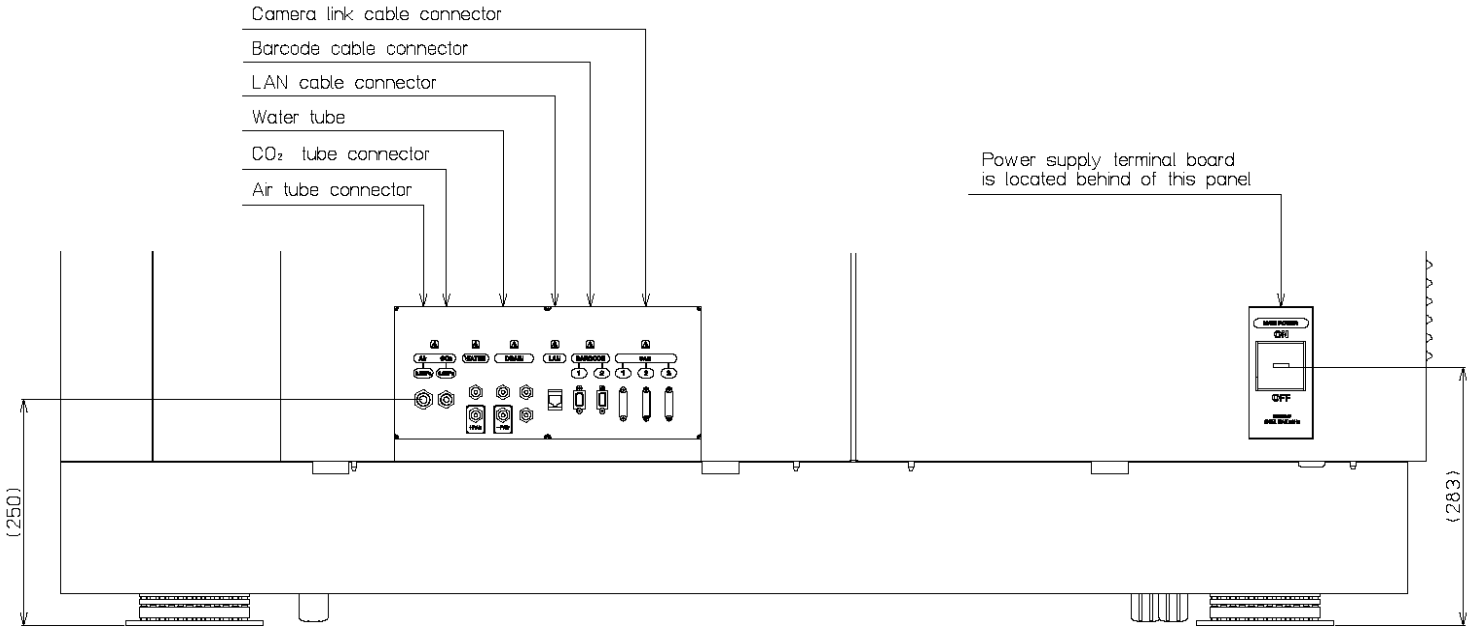
Wiring the Peripherals

Connect the cables after confirming that the power switch on the equipment is turned off.



Connecter boards are located lower-left side of rear panel. Identification of connectors is shown as below.





⚠ WARNING

- Pulling a cable wired to this equipment with a strong force may cause damage to the cable or connected terminals on the equipment. Provide each cable with an ample allowance so that the input terminals on the equipment will not receive a direct pulling force.
- Connect the gas cylinder($CO_2$100%) firmly so that CO_2 will not leak. Install an alarm system as a precautionary measure.

Connection Procedure

Connecting the input power supply to the main unit

1) Remove the rear bottom cover of the equipment.
2) Remove the rear right cover of the equipment.
3) Remove the cover on the input terminals.
4) Connect a power cable (AWG16 or larger that can accommodate 230 VAC, 10 A is recommended) to the input terminals L, N and ⏚. Guide the power cable on the floor and through the power cable wiring aperture in the breaker assembly to connect to the input terminals. As for termination, attach at the end of each lead a crimp terminal or other appropriate terminal matching the terminal screw (M5) and securely connect it to the input terminal.
5) Guide cable clamps through the service apertures and clamp the power cable.
6) After the power cable has been connected, install the cover on the input terminals.
7) Install the rear right cover of the equipment.
8) Install the rear bottom cover of the equipment.

⚠ CAUTION

- The power cable is not supplied. The customer must prepare an appropriate cable in accordance with National Wiring Regulations.
- Be sure to clamp the cable. If cable clamps are not used, the input terminals may receive a load and become loose, resulting in heating of the connection points, electric shock or other dangerous situation.
- The above procedure must be carried out by a qualified electrician.

9) Turn off the switch of the power distribution panel.
10) Connect the power cable to L, N and ⏚ on the power distribution panel. As for termination, attach at the end of each lead a crimp terminal or other appropriate terminal matching the terminal screw on the power distribution panel and securely connect it to the terminal on the panel.

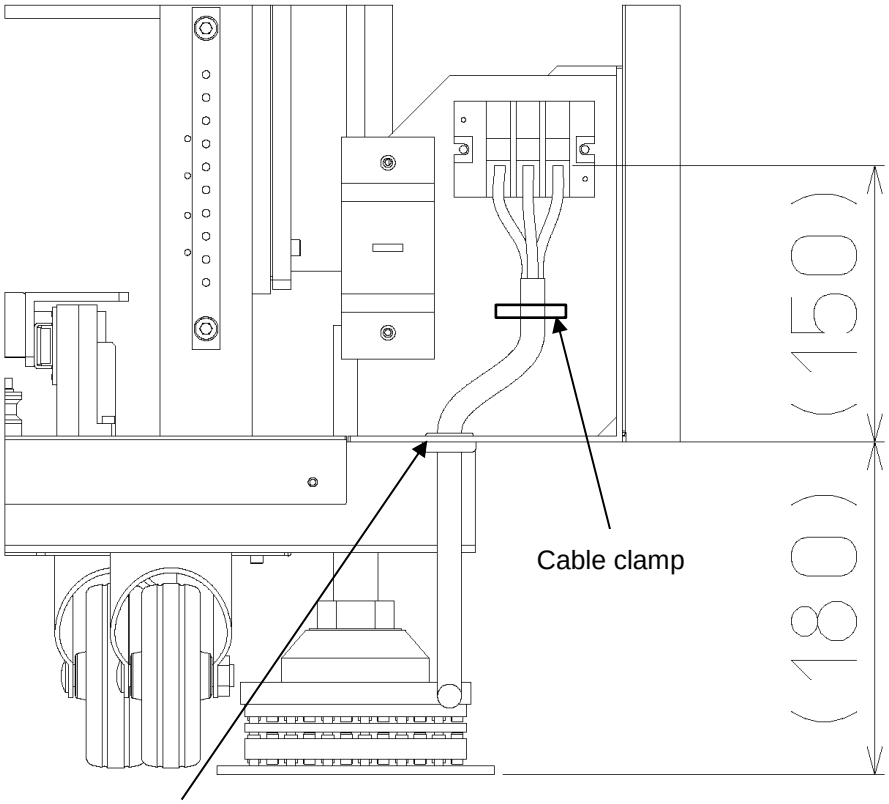
⚠ CAUTION

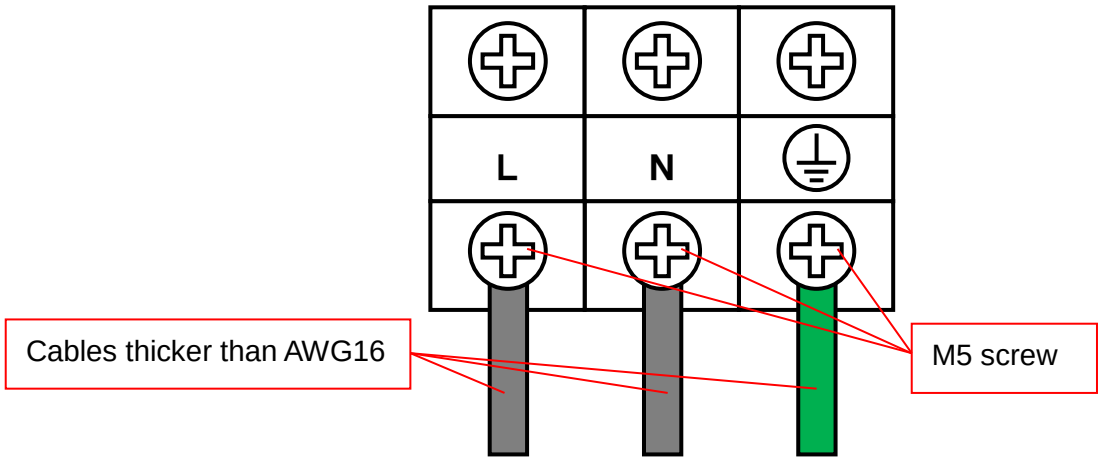
- If the polarities on the power distribution panel are not clear, always ask a qualified electrician or licensed electrician to check.
- Be sure to use the switch of the power distribution panel. If the power distribution panel has no switch, add a switch.
- If the terminal screws are not tightened firmly, the cables may come off or connection points may be heated and create a dangerous situation.
- Be sure to use cables and crimp terminals of matching sizes.

⚠ WARNING

- THIS EQUIPMENT MUST BE EARTH.

Rated power supply voltage : 230VAC
Allowable power-supply voltage fluctuation range : 207 to 253VAC
Rated power frequency : 50Hz
Maximum power consumption : 2kVA

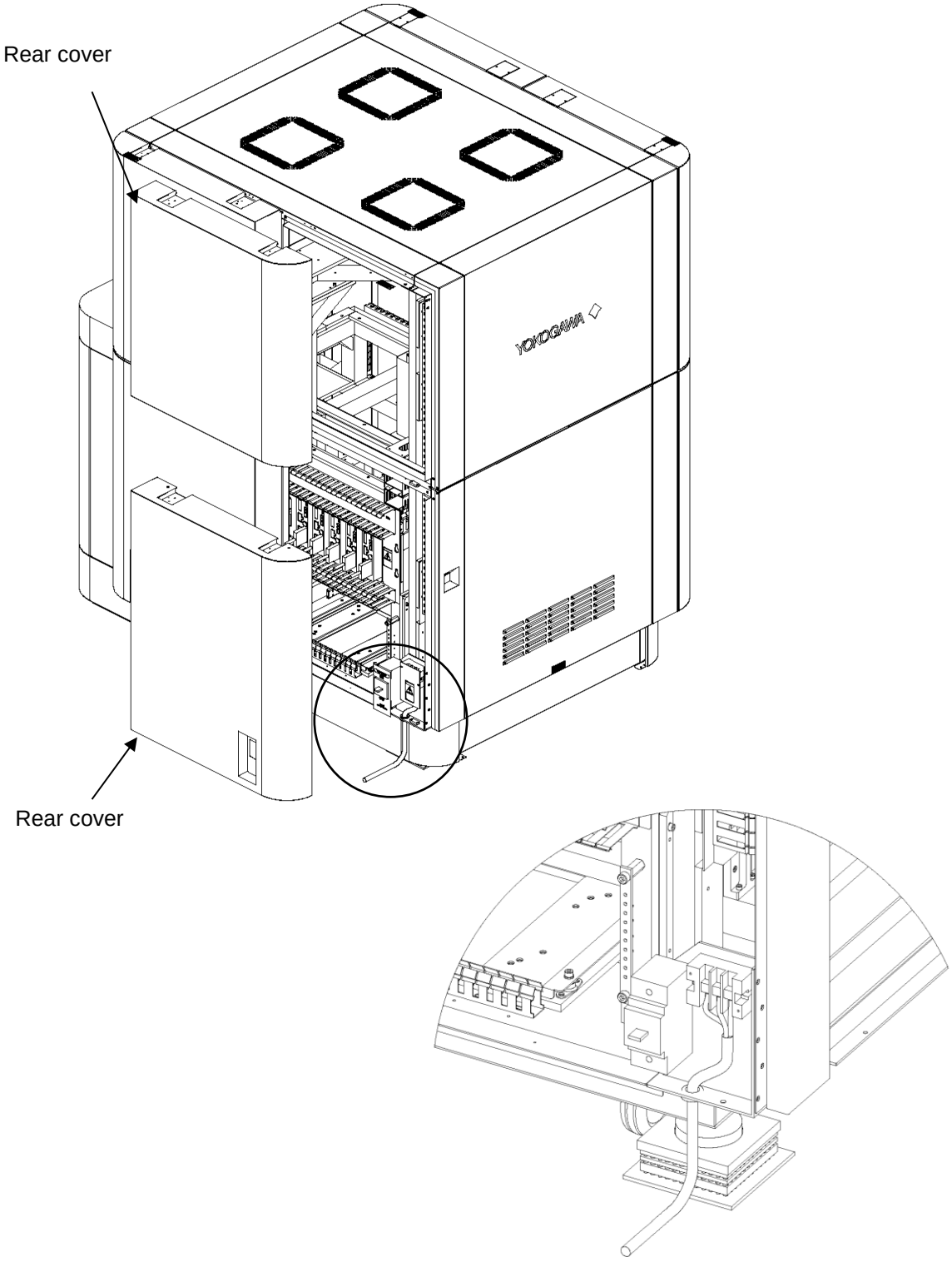




Input terminals
(Rear view)

Parenthetic numbers represent reference dimensions.

Cable Installation



Removal of covers

Before connecting the power, read the following warnings. A failure to observe these warnings may result in electric shock or equipment damage.

⚠ WARNING

●Power supply
Be sure to turn on the power to the equipment after confirming that the power-supply voltage of the equipment matches the voltage of the supplied power.

●Protective grounding
To prevent electric shock, be sure to provide protective grounding before turning on the power to this equipment.

●Defects in protective functions
Do not cut the protective grounding wires running inside or outside this equipment or remove the connected wires from the protective grounding terminals. Doing so will put this equipment in a dangerous condition. Also check the protective functions to ensure absence of defects before operating this equipment.

●Use in gas
Do not operate this equipment in a location where it may come in contact with flammable or explosive gases or vapors. Using this equipment in such environment is very dangerous.

●Removing the case
Only Yokogawa's service personnel can remove the case. This equipment houses high-voltage parts and wires as well as a Class 3B laser, so it is dangerous to remove the case without due caution.

●External connections
Connect each external device after confirming proper protective grounding.

Power Connection to the Workstation and Display

With both, connect the power cable after confirming that the power switch is turned off. Connect the plug on the other end of the power cable to a power outlet meeting the conditions specified below. For the power outlet, use a 3-pin power socket with protective grounding terminal.

Workstation
Rated voltage：230 VAC
Rated current：12 A
Power-supply voltage fluctuation range：207 to 253 VAC
Rated power frequency：50/60 Hz

Display
Rated voltage：230 VAC
Rated current：2 A
Power-supply voltage fluctuation range：207 to 253 VAC
Rated power frequency：50 Hz

Before connecting the power, read the following warnings. A failure to observe these cautions may result in electric shock or equipment damage.

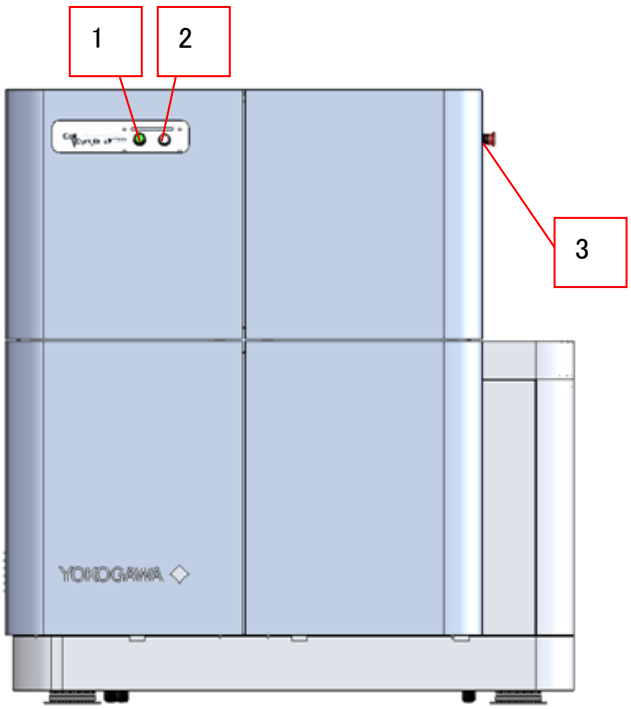
⚠ CAUTION

- Connect the power cable after confirming that the supply voltage matches the rated power-supply voltage of this equipment.
- Connect the power cable after confirming that the power switch on this equipment is turned off.
- To prevent electric shock and fire, be sure to use the power cable supplied by Yokogawa.
- Be sure to provide protective grounding to prevent electric shock. Connect the power cable of this equipment to a 3-pin power outlet with protective grounding terminal. To connect the power cable to a 2-pin power outlet, use a 3-pin to 2-pin conversion adapter (usable only in Japan) and firmly connect the grounding wire of the conversion adapter to the protective grounding terminal of the power outlet.
- Do not use any extension cable without protective grounding wire. Use of such cable will disable the protective operations.

3. Starting and Shutting Down the Equipment

3.1. Name and Function of Each Part

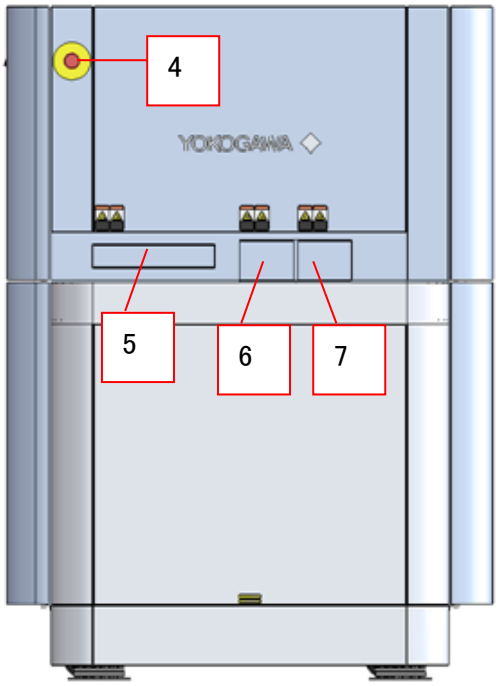
Front View



1) POWER ON button
   Use this button to start the system.
2) POWER OFF button
   Use this button to shut down the system.
3) EMERGENCY STOP button
   Use this button to stop the CV7000 immediately in case of emergency.

Right Side Panel

No barcode reader model



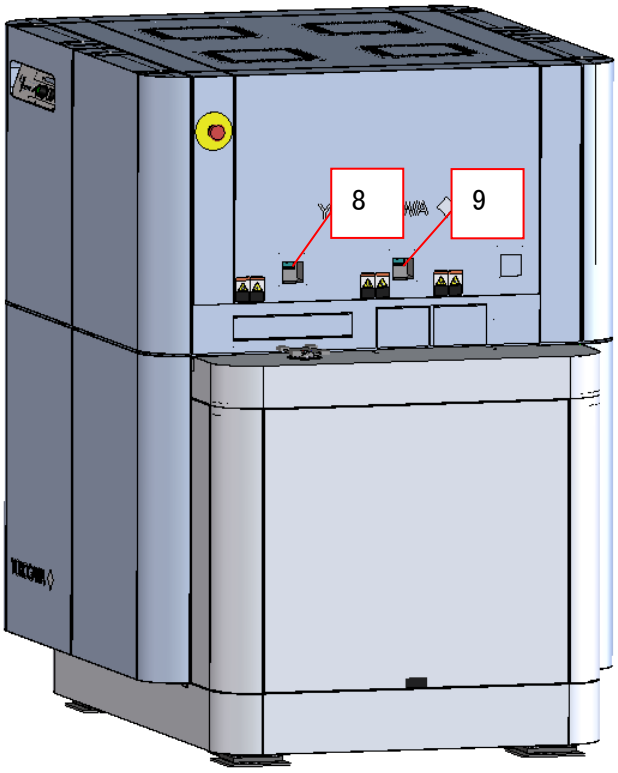
4) EMERGENCY STOP button
Use this button to stop the CV7000 immediately in case of emergency.

5) Assay plate loader
Use this to load the assay plate.

6) Source plate loader
Use this to load the source plate.

7) Tip rack loader
Use this to load a tip rack.

2-barcode reader model

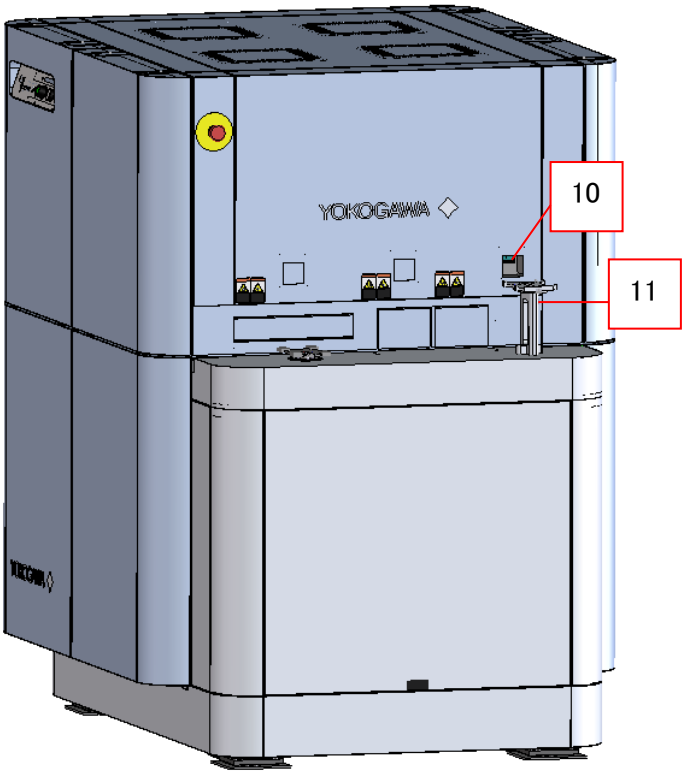


8) Barcode reader for assay plate
Use this to read barcode for the assay plate.
9) Barcode reader for source plate
Use this to read barcode for the source plate.

⚠ WARNING

- Class 1 laser is output from barcode reader. Do not stare into the beam.
- Do not disassemble barcode reader product. Laser emission from this product is not automatically stopped when it is disassembled.

1-barcode reader model



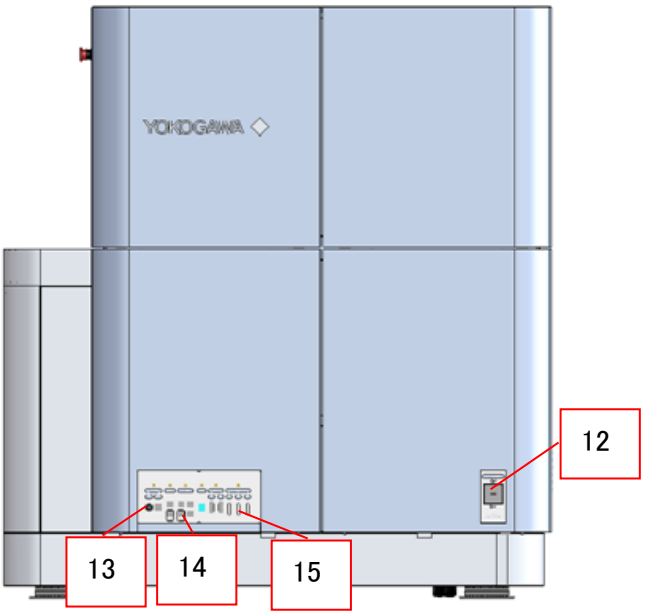
10) Barcode reader for assay plate and source plate
Use this to read barcodes for assay plate and source plate.

11) Reserve space
Use this to put on assay plate and source plate when reading the barcodes.

⚠ WARNING

- Class 1 laser is output from barcode reader. Do not stare into the beam.
- Do not disassemble barcode reader product. Laser emission from this product is not automatically stopped when it is disassembled.

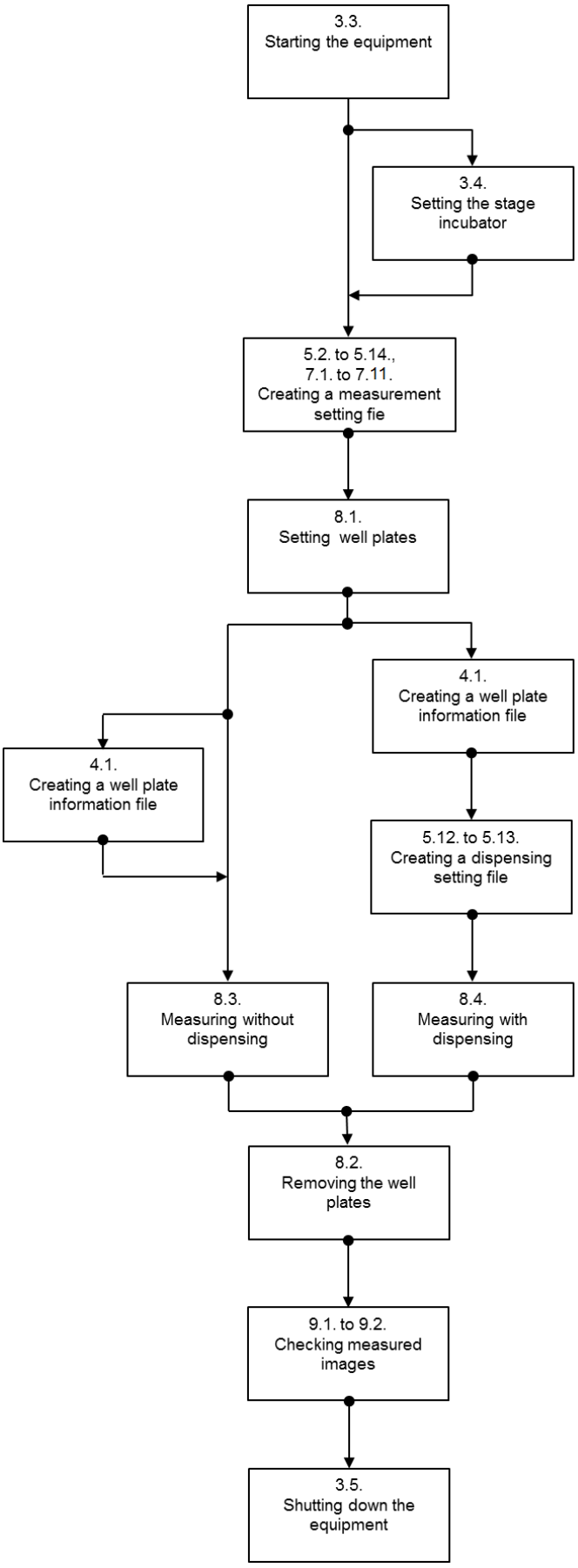
Rear View



12) MAIN POWER breaker
For the main power.
13) Air connector and water connector
Air and water are supplied through these connectors.
14) LAN port
Connect to the system and workstation.
15) Camera port
Connect the camera between the system and workstation.

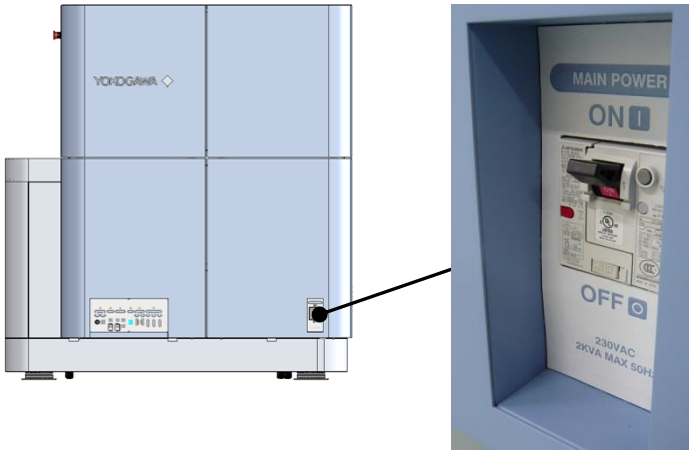
3.2. Flowchart from Equipment Startup to Measurement

A flowchart from equipment startup to measurement is shown below.

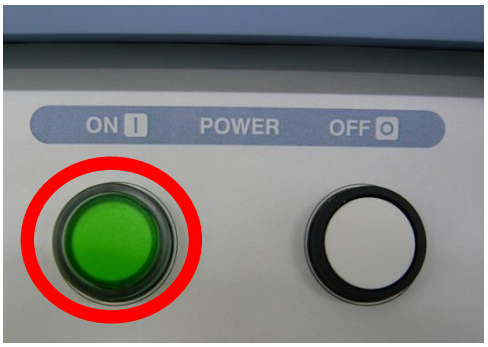


3.3. Starting the Equipment

1) Turn on the MAIN POWER breaker on the rear side of the equipment.



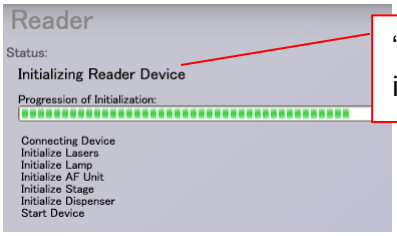
2) Press the POWER ON button on the front side of the equipment. The POWER ON lamp becomes lit.



3) Start the measurement PC.

4) Click the icon below on the desktop of the measurement PC to start the application software.

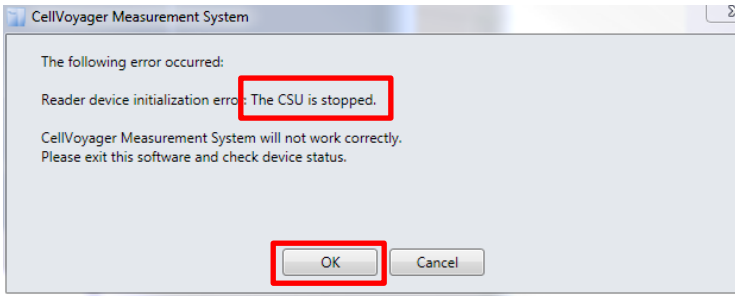
5) The portal application software starts. Wait a while until the start is completed.



"Ready" appears when the start is completed.

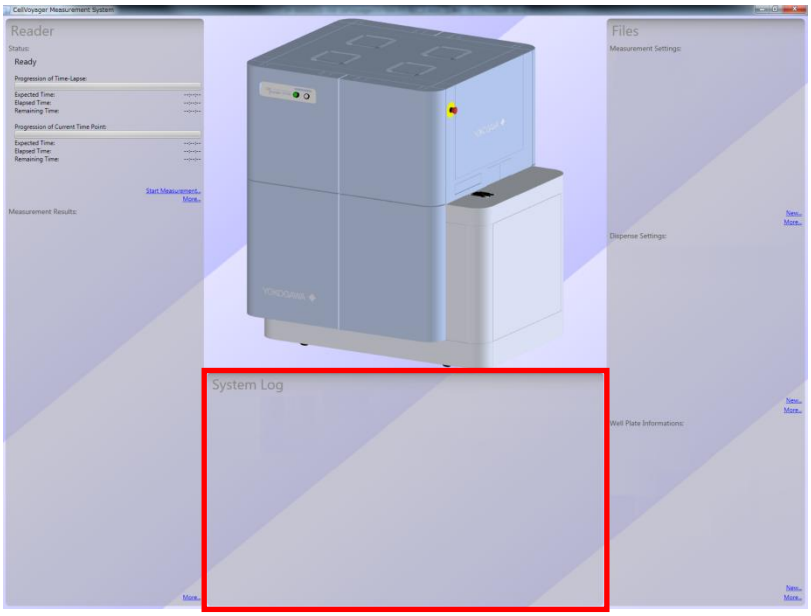
MEMO

●In case that following window appears and it says “The CSU is stopped”, click “OK” (after clicking application software closes automatically).



After application software closes, please re-start CV7000 (press POWER OFF button to shut down, and press POWER ON button to turn on).

If the ⊗ mark appears in the System Log field, stop measurement and contact us.



Refer to 3.6 for the screen of the application software.

3.4. Setting the Stage Incubator

The stage incubator is set. (Stage incubator model only)

MEMO

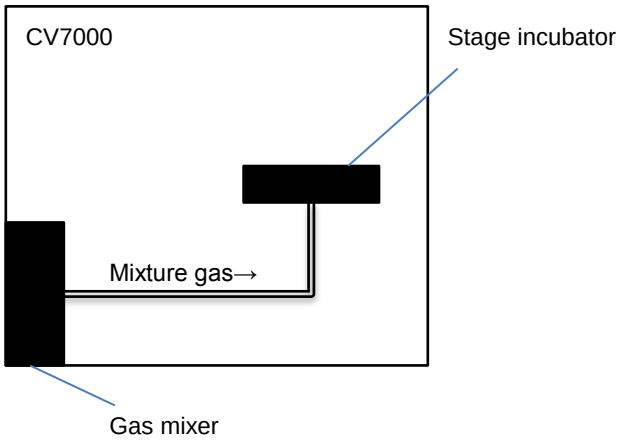
●CO_2 density control is different in CV7000 which is sold before March 2015 (conventional model) and after April 2015 (CV7000*S*)

- Conventional model (CV7000)

  Set CO_2 density and gas flow rate at outlet of gas mixer

- CV7000*S*

  Set CO_2 density at stage incubator. Gas flow rate is fixed to optimal value (not settable)



●Hardware of CV7000 must be altered to change the CO_2 density control from conventional model to CV7000*S* (paid service)

Please contact dealer for detail

Preparations

1) Confirm the connection of the CO_2 cables.

2) Install a CO_2 detector to avoid carbon-dioxide toxicity.

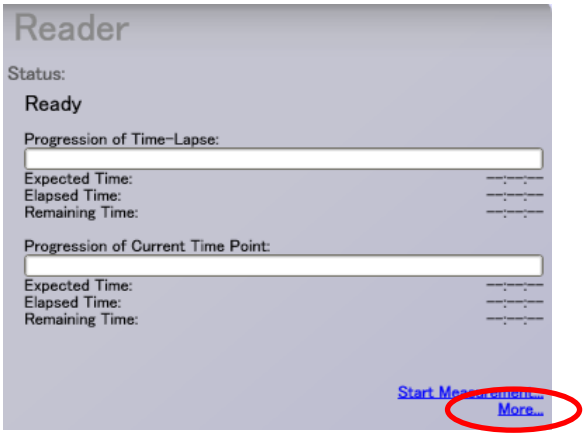
3) Prepare the water supply bottle and the drainage bottle. Put pure water into the water supply bottle. (Pour off the water in the bottles regularly.)

⚠ WARNING

- In the case of replacing water in the bottles, make sure that the MAIN POWER breaker surely turns OFF to shut down CV7000. (Refer to 3.5) Be careful not to turn the breaker ON by oversight while at work.

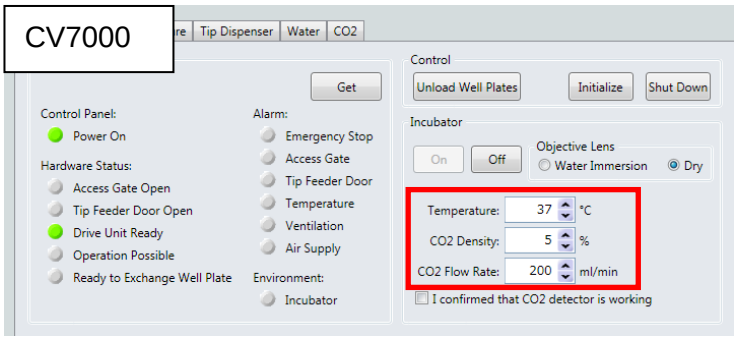
Starting Stage Incubator

1) Click “More” at the bottom of the Status area on the Reader screen.

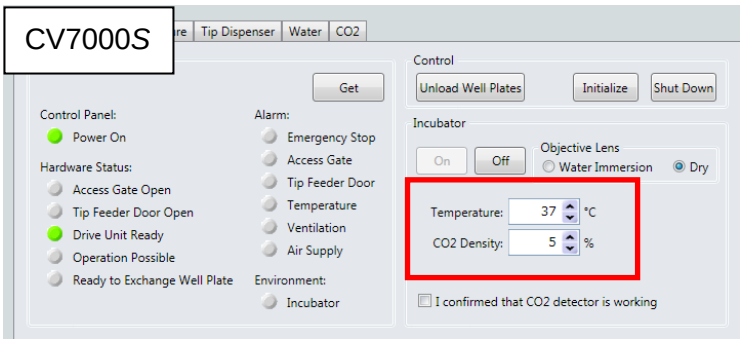


2) The “Device Console” tab opens in the top left of the “Reader Control” screen. (Refer to 6.4)
Input temperature, CO_2 density and gas flow rate (Only CV7000).

CV7000



CV7000*S*

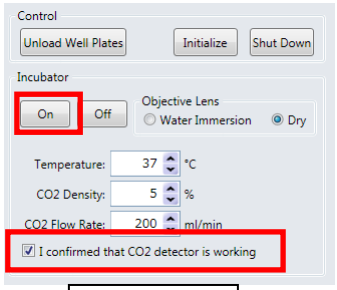


MEMO

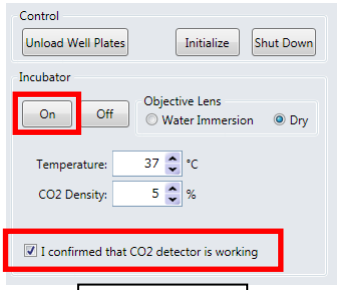
●Settable range of CO_2 density and gas flow rate is as following.

- CV7000
  Settable range of CO_2 density: 0.0～9.9%
  Settable range of gas flow rate: 0～500ml/min
- CV7000*S*
  Settable range of CO_2 denisty: 4.5～5.5%
  Settable range of gas flow rate: Unsettable
  (fixed to optimal value)

3) Confirm that the CO_2 detector is operating. And then, check the “I confirmed that CO2 detector is working” checkbox.

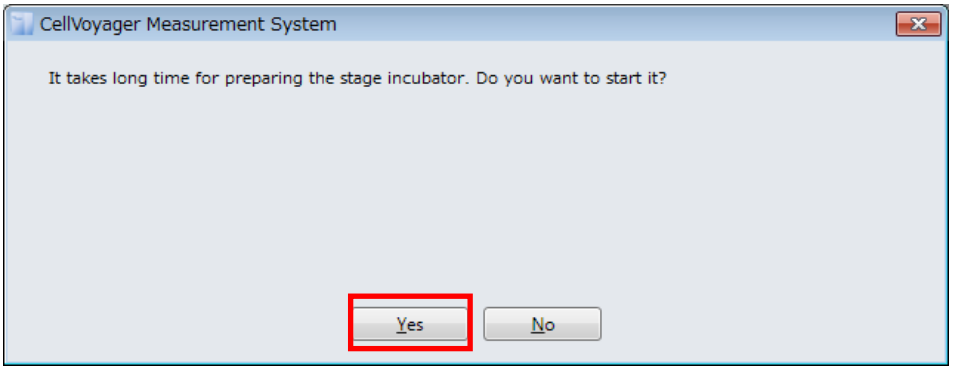


CV7000

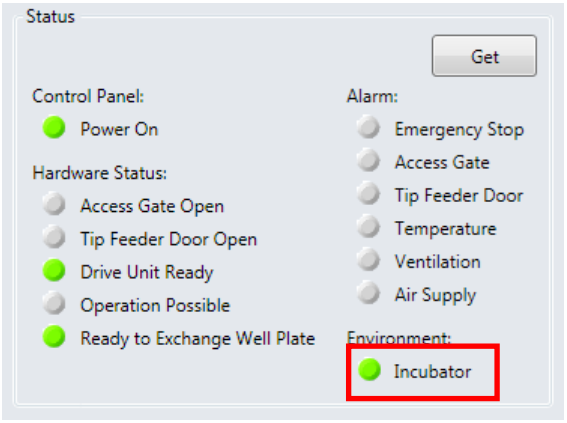


CV7000*S*

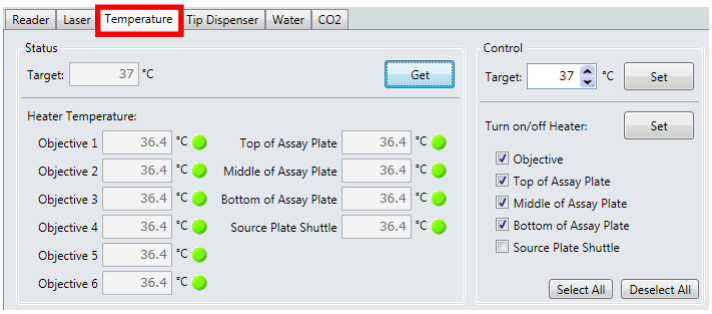
4) In case that following diagram is shown. Click “OK” and Stage Incubator preparation starts. (It takes about 10 minutes.)

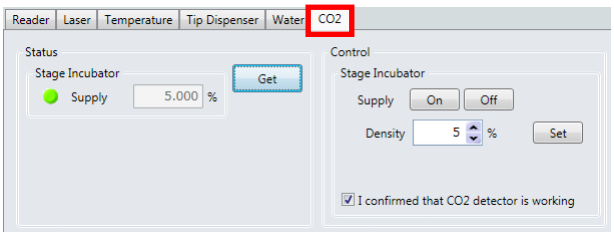


5) Indicator of “Incubator” becomes green after water supplying to Stage Incubator finishes and temperature/ CO_2 density become stable. (It takes about 40 minutes.)



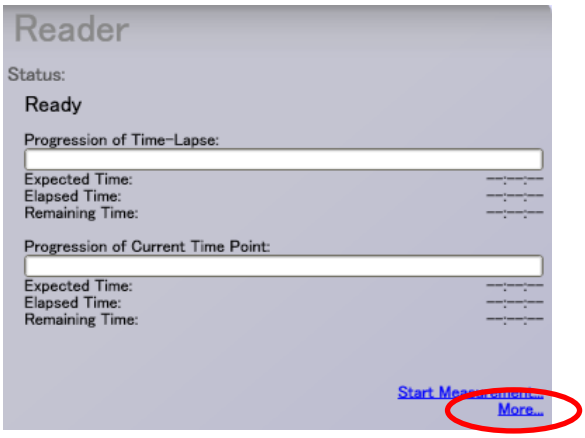
6) Present temperature and CO_2 density can be confirmed by selecting “Temperature” tab and “CO2” tab.





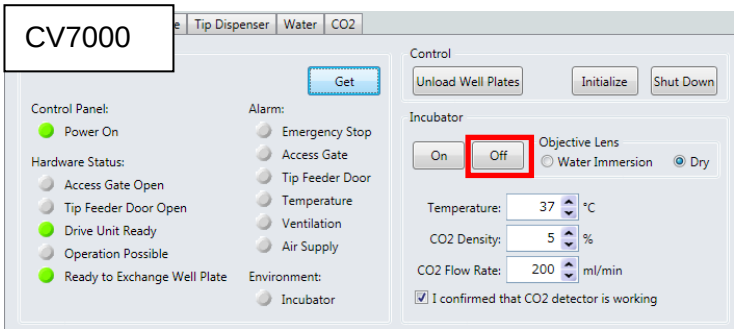
Finishing Stage Incubator

1) Click “More” at the bottom of the Status area on the Reader screen.

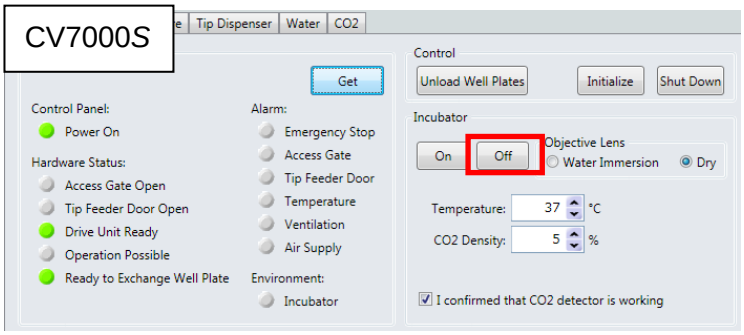


2) The “Device Console” tab opens in the top left of the “Reader Control” screen. (Refer to 6.4)
Click “OFF”.

CV7000



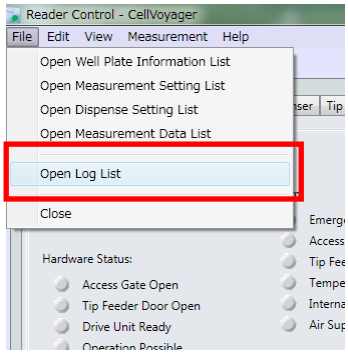
CV7000S



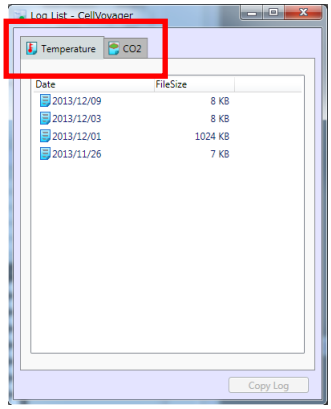
Confirming Log File for Stage Incubator

Each log file of temperature and CO2 concentration for stage incubator can be output.

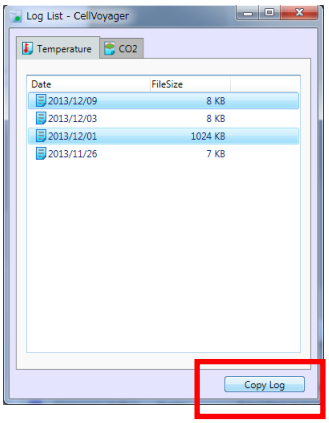
1) To open log files for temperature and CO2 concentration, Select the “File” menu of the “Reader Control” screen and then click “Open Log List.”



(2) The “Log List” screen is shown.
Select the “Temperature” tab or the “CO2” tab.

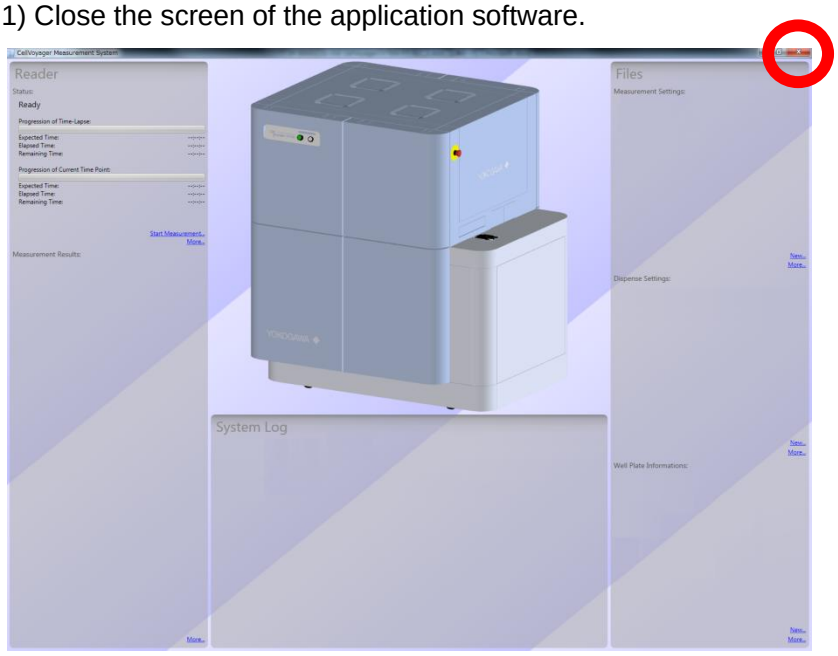


(3) Select the items you desire and click “Copy Log” to select destination to be output.



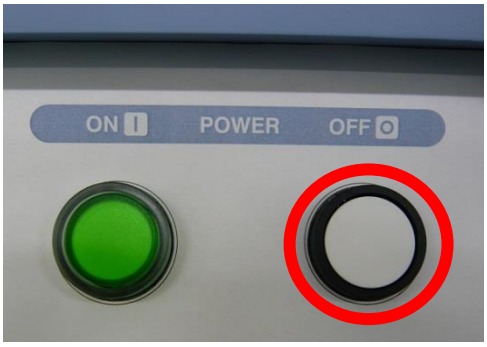
3.5. Shutting Down the Equipment

1) Close the screen of the application software.



2) Turn off the power of the measurement PC.

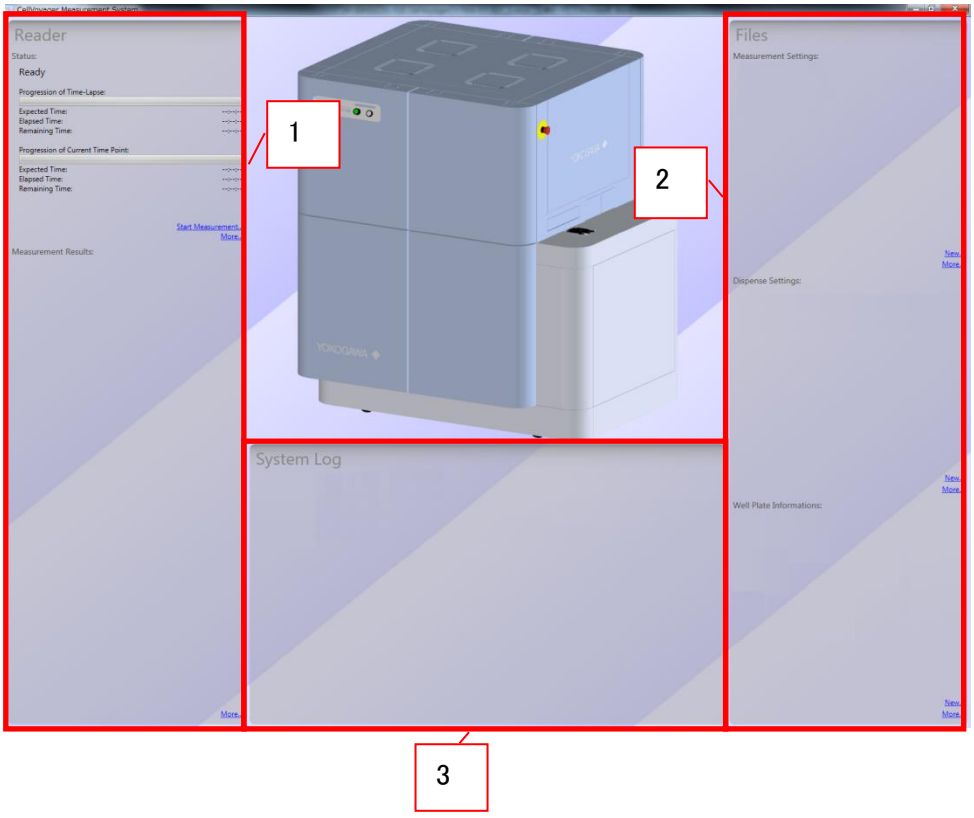
3) Press the POWER OFF button.
The POWER ON lamp turns off.



4) Turn off the MAIN POWER breaker on the rear side of the equipment.

3.6. Explanation of the Main Screen

The main screen of the portal application software is divided into the Reader area, Files area and log information display area.



1) Reader area (Refer to 6.1)
Well plates are set up for imaging and measured.

2) Files area (Refer to 6.1)
The past imaging history, dispenser settings and history of well plate information files are shown.

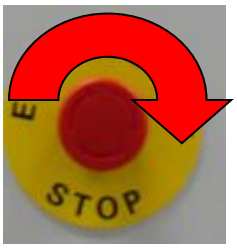
3) Log information display
System logs are shown.

3.7. In Case of Emergency

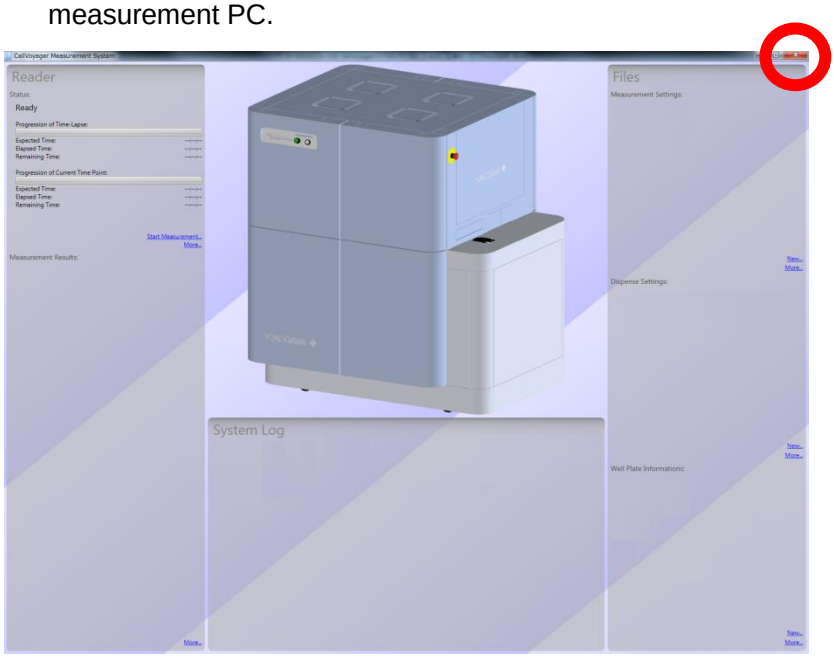
This section explains how to stop the equipment immediately in case of emergency and how to reinitialize CV7000.

1) Press the EMERGENCY STOP button.

2) To release the EMERGENCY STOP button, turn it clockwise.



3) Close the application software. Additionally, turn off the power of the measurement PC.



4) Turn off the MAIN POWER breaker on the rear side of the equipment.

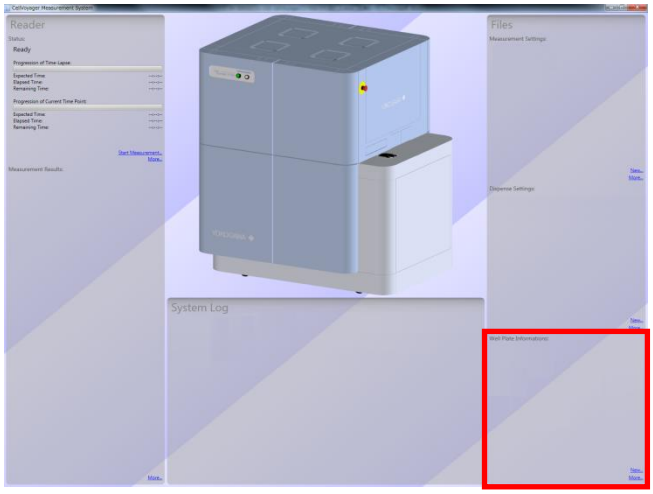
5) After 10 seconds, start CV7000. (Refer to 3.3.)

4. Entering Well Plate Information Files

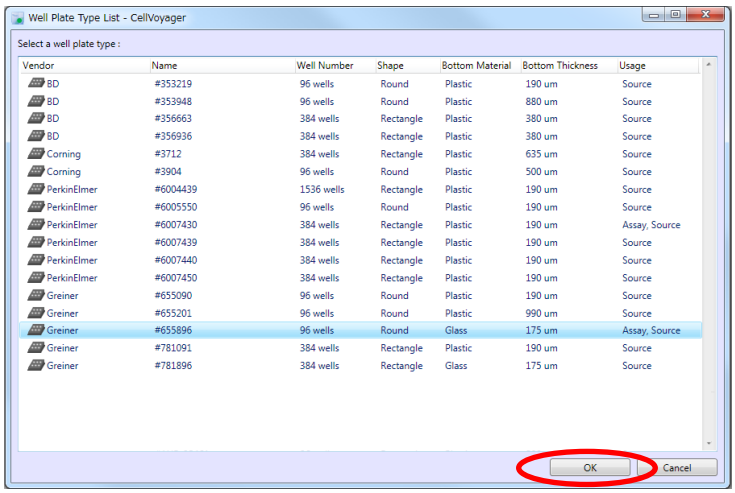
It is not necessary to enter the well plate information files for measuring. (Entry is recommended.)
But if dispensing is performed, this file must be created.

4.1. Creating a Well Plate Information File

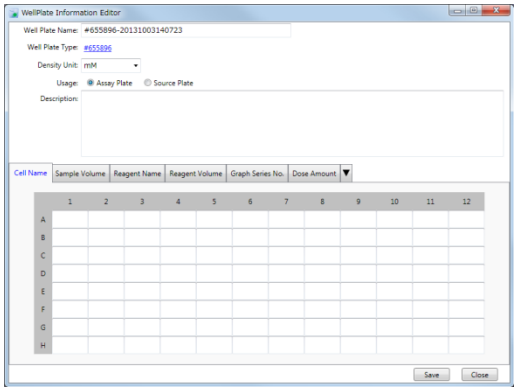
1) Click “New” at the bottom of the Well Plate Informations area.



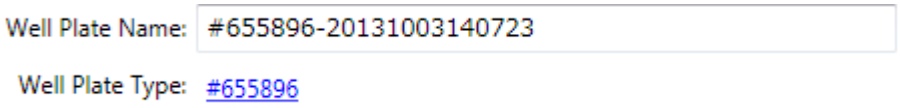
2) Select a desired well plate product. After a product has been selected, click “OK.” (Refer to 4.3)



The screen for entering well plate information opens. (Refer to 4.3)



3) Enter the name of the well plate (file name).



4) Select “Usage.”
Select “Assay Plate” if the well plate contains a cell sample to be measured.
Select “Source Plate” if the well plate contains a reagent or other compound.

5) Enter information regarding the well. (Refer to 4.2)

Displayed Well Information Items

Item	Explanation	Remarks
Cell Name	Cell name	∗1
Sample Volume	Amount of solution in the assay plate well (µl)	∗2
Reagent Name	Reagent name. After imaging analysis, a density-dependent curve will be the drawn based on the reagent name entered here.	∗3
Reagent Volume	Amount of solution in the source plate well (µl)	∗2
Graph Series No.	Graph number. When density-dependent curves are drawn after imaging analysis, wells of the same number are reflected as data points on the same graph.	∗4
Dose Amount	Density of reagent in the assay plate. This information is reflected in the X-axis of the density-dependent curve after imaging analysis.	∗4

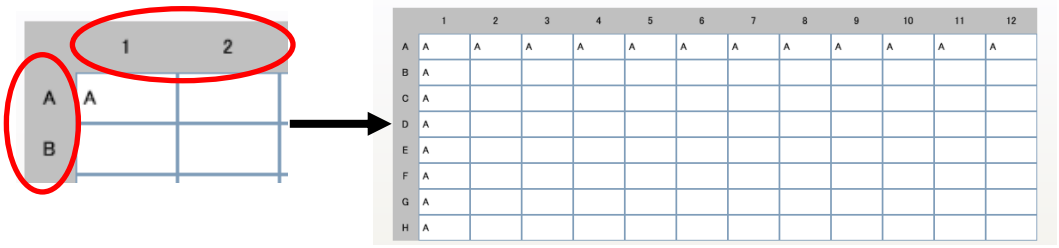
∗1 : This information is not reflected in measurement or imaging analysis. (The field may remain blank.)
∗2 : This information must be entered if dispensing is performed.
∗3 : This information is displayed on the density-dependent curve.
(Entry is recommended if CV7000 Analysis Software is installed.)
∗4 : This information must be entered properly to draw a desired density-dependent curve. (Entry is recommended if CV7000 Analysis Software is installed.)

If the “Dose Amount” field is entered, select “Density Unit.” (You can also use the default unit without entering a specific unit.)

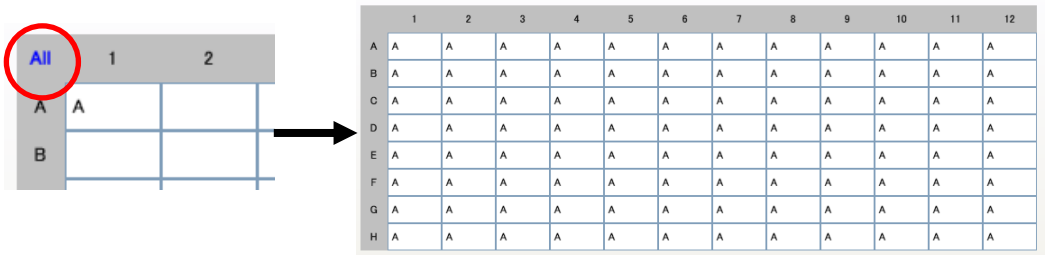
6) Click “Save.”

4.2. How to Enter Well Information

Click the number or alphabet letter of the well to enter the number, character, or empty space at the beginning of the row or column of the well in the entire row or column.



Place the mouse pointer over the left upper corner of the well to display “All”. Click to enter the number, character, or empty space you entered in cell A1 in all the remaining cells.



You can copy and paste the data you enter in the Excel worksheet.

Data entered in Excel worksheet

	1	2	3	4	5	6	7	8	9	10	11	12
A	0	1	3	10	30	100	300	1000	3000	10000	30000	100000
B	0	1	3	10	30	100	300	1000	3000	10000	30000	100000
C	0	1	3	10	30	100	300	1000	3000	10000	30000	100000
D	0	1	3	10	30	100	300	1000	3000	10000	30000	100000
E	0	1	3	10	30	100	300	1000	3000	10000	30000	100000
F	0	1	3	10	30	100	300	1000	3000	10000	30000	100000
G	0	1	3	10	30	100	300	1000	3000	10000	30000	100000
H	0	1	3	10	30	100	300	1000	3000	10000	30000	100000

Paste

	1	2	3	4	5	6	7	8	9	10	11	12
A	0	1	3	10	30	100	300	1000	3000	10000	30000	100000
B	0	1	3	10	30	100	300	1000	3000	10000	30000	100000
C	0	1	3	10	30	100	300	1000	3000	10000	30000	100000
D	0	1	3	10	30	100	300	1000	3000	10000	30000	100000
E	0	1	3	10	30	100	300	1000	3000	10000	30000	100000
F	0	1	3	10	30	100	300	1000	3000	10000	30000	100000
G	0	1	3	10	30	100	300	1000	3000	10000	30000	100000
H	0	1	3	10	30	100	300	1000	3000	10000	30000	100000

The following shows an entry example of well information.

●Entry Example of Reagent Names

A density-dependent curve is output for each reagent name you have entered. Leave the field blank for wells that are not used.

Cell Name | Sample Volume | Reagent Name | Reagent Volume | Graph Series No. | Dose Amount

	1	2	3	4	5	6	7	8	9	10	11	12
A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A
B	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A
C	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A
D	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A	Reagent A
E	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B
F	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B
G	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B
H	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B	Reagent B

●Entry Example of Graph Series Numbers

When density-dependent curves are output after imaging analysis, wells of the same number are displayed as data points on the same graph. Enter “0” for wells that are not used.

Cell Name | Sample Volume | Reagent Name | Reagent Volume | Graph Series No. | Dose Amount

	1	2	3	4	5	6	7	8	9	10	11	12
A	1	1	1	1	1	1	1	1	1	1	1	1
B	2	2	2	2	2	2	2	2	2	2	2	2
C	3	3	3	3	3	3	3	3	3	3	3	3
D	4	4	4	4	4	4	4	4	4	4	4	4
E	5	5	5	5	5	5	5	5	5	5	5	5
F	6	6	6	6	6	6	6	6	6	6	6	6
G	7	7	7	7	7	7	7	7	7	7	7	7
H	8	8	8	8	8	8	8	8	8	8	8	8

●Entry Example of Dose Amounts

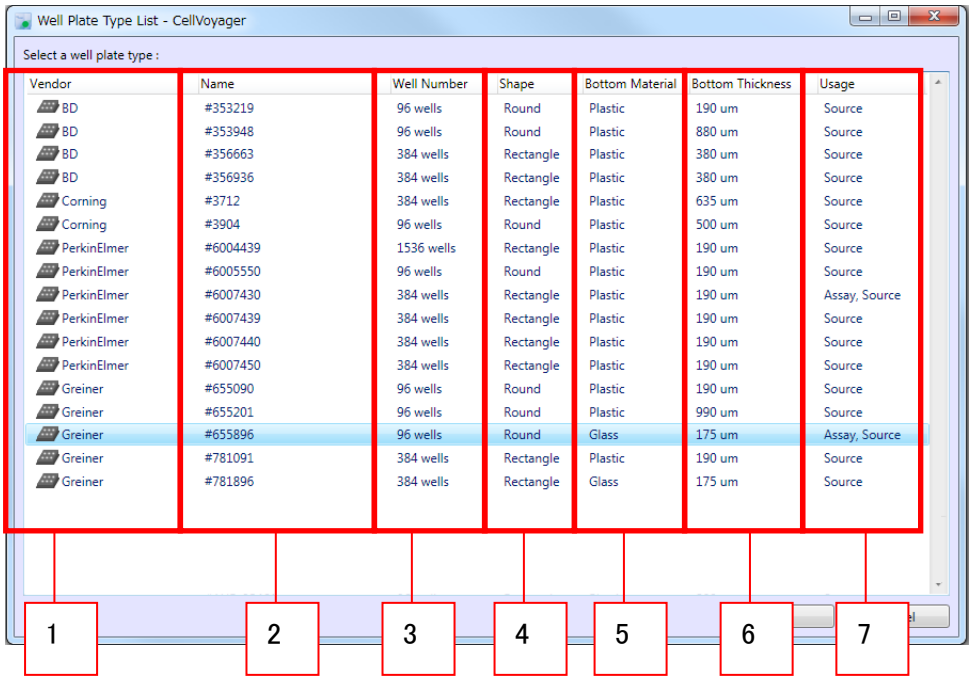
When density-dependent curves are output after imaging analysis, this information is reflected in the X-axis of each curve. Enter “0” for wells to which no reagent was added or wells that are not used.

Cell Name | Sample Volume | Reagent Name | Reagent Volume | Graph Series No. | Dose Amount

	1	2	3	4	5	6	7	8	9	10	11	12
A	0.01	0.03	0.06	0.1	0.3	0.6	1	3	6	10	30	100
B	0.01	0.03	0.06	0.1	0.3	0.6	1	3	6	10	30	100
C	0.01	0.03	0.06	0.1	0.3	0.6	1	3	6	10	30	100
D	0.01	0.03	0.06	0.1	0.3	0.6	1	3	6	10	30	100
E	0.01	0.03	0.06	0.1	0.3	0.6	1	3	6	10	30	100
F	0.01	0.03	0.06	0.1	0.3	0.6	1	3	6	10	30	100
G	0.01	0.03	0.06	0.1	0.3	0.6	1	3	6	10	30	100
H	0.01	0.03	0.06	0.1	0.3	0.6	1	3	6	10	30	100

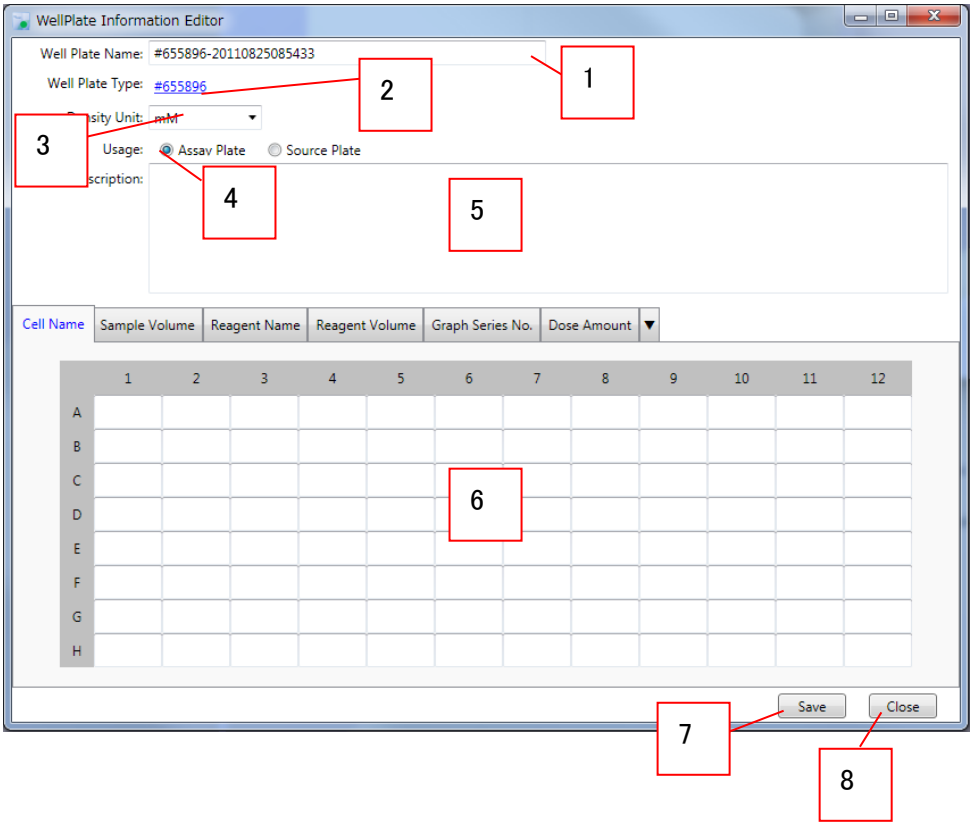
4.3. Well Plate Information File Screen

Well Plate Product Selection Screen

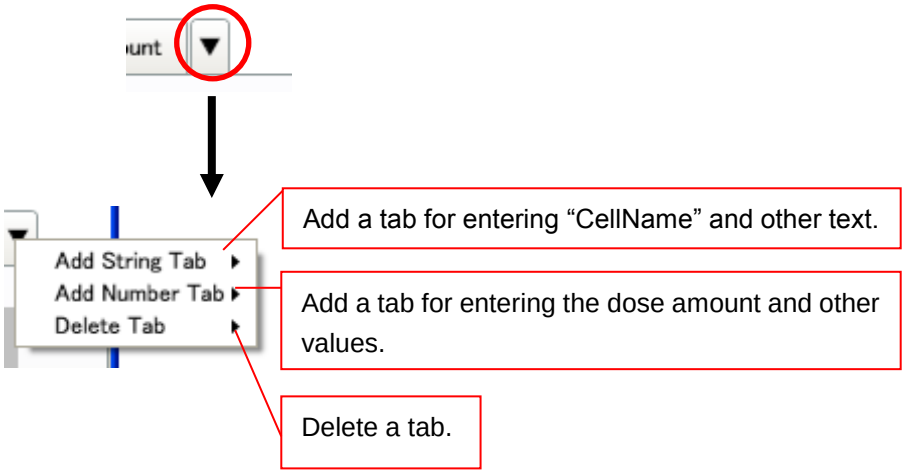


1) Well plate manufacturer
2) Well plate type (model number)
3) Number of wells
4) Well shape
5) Material of well plate bottom
6) Thickness of well plate bottom
7) Well plate purpose

Well Plate Information File Screen



1) Well plate information file name
2) Well plate type
   (Clicking allows you to replace with other same well-number plate.)
3) Unit of dose amount (density of reagent in the assay plate)
4) Selection of well plate purpose (assay plate/source plate)
5) Memo
6) Well information
   Selecting ▼ at the right edge of the tab opens the menu where you can add tabs for entering well information.



7) Save the well plate information file.
8) Close the well plate information file screen.

5. Using the Measurement Software

5.1. Measurement Software Functions

Time-lapse Setting

You can set a timeline to be used as the basis of measurement operation and processing or set time-lapse measurement for each plate. In time-lapse measurement for each plate, imaging of the same well plate is repeated at the specified interval.
You can add as many fluorescence imaging, software focus and other processes as desired for each timeline.

Well Plate Scan Setting

You can set an imaging well and imaging points.
To set an imaging well, select the assay plate well to be measured and save the setting information. To set imaging points, select imaging points in the well and movement pattern, and save the setting information. Imaging points are set in the following five modes.

●Cell Count Function

The system moves through the imaging points to capture images repeatedly in the same well until the specified cell count is reached. When the number of repetitions reaches the specified value, imaging will stop and the system will move to the next well.

●Cell Search Function

The system moves through the imaging points repeatedly until the specified count is reached, and outputs images of the specified number at the imaging point associated with the largest cell count, or imaging point closest to the specified cell count.

●Tile Function

Images of the whole region of well (excluding edge areas or including edge areas) are captured with the specified object lens and tiled from the one corresponding to the top left of the well. A desired overlap between captured images can be specified in pixels.

●Partial Tile Function

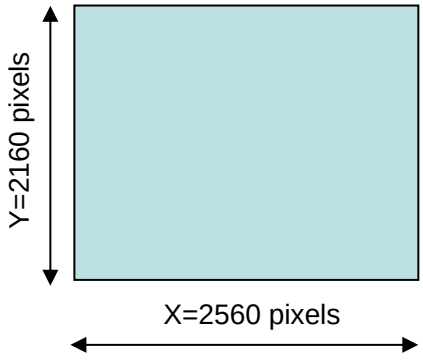
Images of the partial region(s) of well (capable of plural regions setting) are captured with the specified object lens and tiled from the one corresponding to the top left of the well. A desired overlap between captured images can be specified in pixels.

●Fixed Position Function

Positions of imaging points are specified directly. You can specify desired imaging points or imaging points in a circular, rectangular or other pattern.

Magnification factors and corresponding view fields of the camera

Magnification	X	Y
4x	4160μm	3510μm
10x	1664μm	1404μm
20x	832μm	702μm
40x	416μm	351μm
60x	277μm	234μm



View field of the camera in pixels

Action List

You can set various measurement operations including software focus, fluorescence imaging, bright field/phase contrast imaging, 3D imaging and dispensing.

Software Focus (Imaging Auto-focus)

Images are captured repeatedly at a specified wavelength while moving the Z position on the confocal plane to determine the Z position on the confocal plane at the position of the brightest image. The software focus setting has no imaging function. The software focus function is inter-dependent with fluorescence imaging and 3D imaging, so when the software focus function was used, you can perform fluorescence imaging and 3D imaging successively from the condition of the software focus function being executed.

Fluorescence Acquisition

Fluorescent light from the target is captured. Multiple cameras are used to capture images simultaneously at one or more fluorescent wavelengths. To acquire images for each wavelength during measurement where two or more wavelengths are used, add two or more fluorescence imaging sessions and select a desired measurement wavelength in each session. You can capture soft focus images by setting fluorescence imaging immediately after

implementing the software focus function. If software focus is skipped and only fluorescence imaging is set, auto-focus imaging can be performed.

●Auto-focus Imaging
One shot imaging is performed without software focus.

●Software focus imaging
One shot imaging is performed with software focus.

●Epifluorescence Imaging
An optical circuit detouring around the confocal unit is used. The laser is used as the light source to capture images in the epifluorescence mode.

●High-speed Time-lapse Imaging
Time-lapse measurement is performed for each well at an interval of several tens to several hundreds of milliseconds. In time-lapse measurement for each well, imaging of the same well is repeated at the specified interval. Time-lapse imaging of one well is completed before imaging of the next well is performed. During high-speed time-lapse imaging, dispensing can be performed at a specified timing. (Dispenser model only)

3D Fluorescence Acquisition

This is a function whereby a Z-position imaging area is specified and fluorescence images are captured over the entire Z plane by moving the Z position on the confocal plane. Fluorescent images with one or more wavelengths are captured. To acquire images for each wavelength during measurement where two or more wavelengths are used, add two or more 3D imaging sessions and select a desired measurement wavelength in each session. You can perform 3D imaging using the software focus plane as the reference, by setting 3D imaging immediately after implementing the software focus function. If software focus is skipped and only 3D imaging is set, 3D imaging can be performed using the auto-focus plane as the reference.
In regard to captured 3D images, a single image projected in the Z-axis direction also can be created.

●Maximum
You can perform MIP on captured 3D images. MIP, or Maximum Intensity Projection, is a function to build a single image by putting together pixels associated with the largest signals among identical pixels from multiple image data captured on the confocal plane in the Z-axis direction. This way, an object that cannot fit a single confocal image can be fully depicted by capturing its 3D images and synthesizing them via MIP.

●Minimum
You can perform MinIP on captured 3D images. MinIP, or Minimum Intensity Projection, is a function to create a single image by putting together pixels associated with the smallest signals among identical pixels from multiple

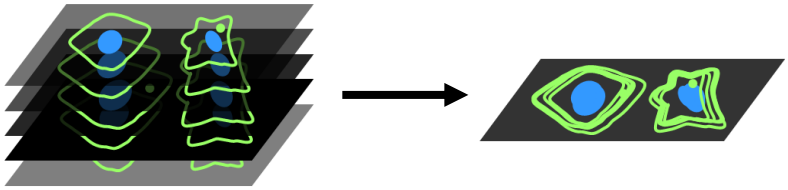
image data captured on the confocal plane in the Z-axis direction. This function is effective for bright-field images captured by 3D imaging.

●Average

You can perform AIP on captured 3D images. AIP, or Average Intensity Projection, is a function to create a single image by putting together pixels associated with the average signals among identical pixels from multiple data captured on the confocal plane in the Z-axis direction.
Average values are obtained by dividing the total pixel intensity by the number of Z images.

●Sum

SUM is a function to create a single image by putting together pixels associated with the summation signals among identical pixels from multiple image data captured on the confocal plane in the Z-axis direction. To calculate summation intensity, the value which subtracted the background of the camera is summed from the 2nd sheet. Imageable intensity is up to 65525.



Z-axis projection

Bright-field/Phase-contrast Acquisition (Bright field/ phase contrast model only)

This is a function whereby imaging is performed based on the bright field and phase contrast by using a lamp as the light source. In the phase contrast mode, the object lens must be changed to one for phase contrast imaging.

●Auto-focus Imaging

One shot imaging is performed without software focus.

●High-speed Time-lapse Imaging

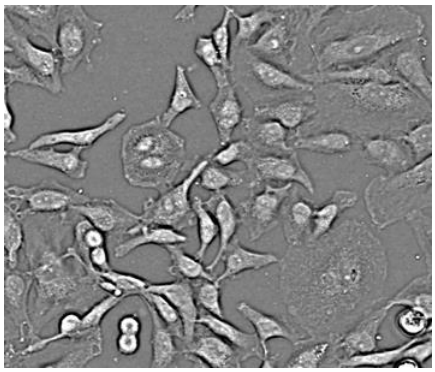
Time-lapse measurement is performed for each well at an interval of several tens to several hundreds of milliseconds. In time-lapse measurement for each well, imaging of the same well is repeated at the specified interval. Time-lapse imaging of one well is completed before imaging of the next well is performed. During high-speed time-lapse imaging, dispensing cannot be performed.

Z-Stack Bright-field/Phase-contrast Acquisition (Bright field/ phase contrast model only)

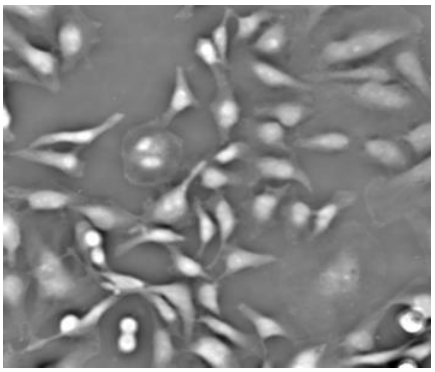
This is a function to capture Z-stack bright field and phase contrast images by moving the Z position on the focal plane. This is captured with other timing separated from fluorescence acquisition. In the phase contrast mode, the object lens must be changed to one for phase contrast imaging.
In regard to captured Z-stack images, a single image projected in the Z-axis direction also can be created in common with 3D Fluorescence Acquisition.

DPC (Digital Phase-contrast) Acquisition (Bright field model only)

This is a function to capture 2 Z position images by using a lamp as the light source and make phase contrast like image (Phase type) or fluorescence like image (Fluor type). This is captured with other timing separated from fluorescence acquisition.



Phase type DPC image



Fluor type DPC image

Dispensing Operation (Dispenser model only)

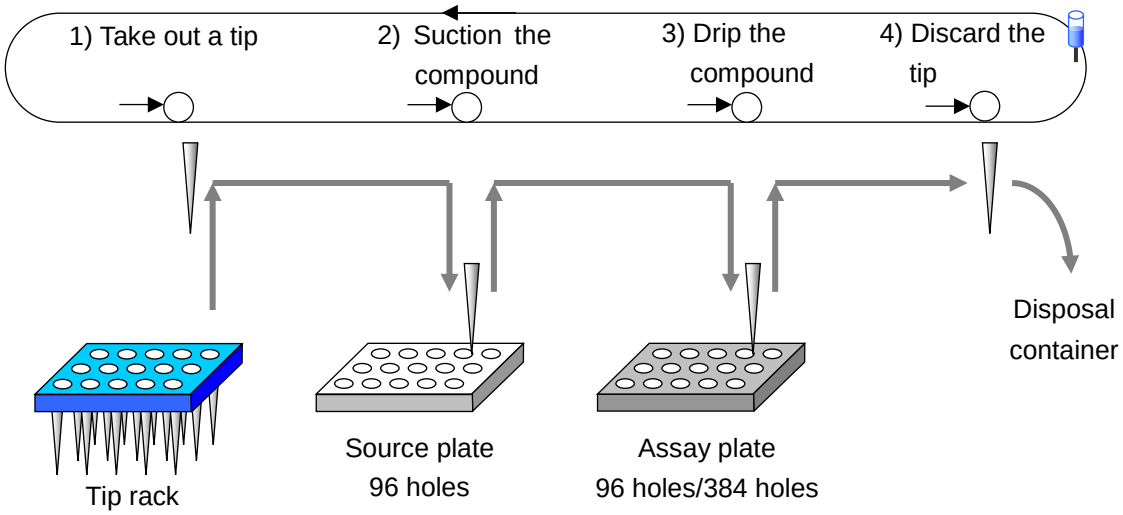
This is a function whereby reagent is dripped using a dispenser. Dispensing is performed for each plate, meaning that reagent is dispensed into all specified wells in one well plate.
Since dispensing is not associated with any imaging function, separate measurement operations must be added if images are to be captured before and after dispensing.

Dispensing Function

Dispensing Mechanism

With the CellVoyager, dispensing refers to the operations such as suctioning a specified volume of a compound from the source plate that contains the compound and dripping the compound onto the assay plate. The device that performs dispensing is called “Dispenser.”

The dispenser of the CellVoyager performs dispensing using disposable tips one by one. After each tip has been used, the dispenser replaces the tip with a new one and performs the next dispensing action. The figure below illustrates the operation flow of the dispenser.



Overview of Dispensing Mechanism

1) Taking out a tip

An unused tip is taken out from the tip rack. After all unused tips on the tip rack have been used, the tip rack is replaced with one containing unused tips.

2) Suctioning the compound

The edge of the tip is moved to above a specified well in the source plate containing the compound, and the compound is suctioned from the well by a specified volume.

3) Dripping the compound

The tip containing the compound is moved to above a specified well in the assay plate, and the compound is dripped into the well.

4) Discarding the tip

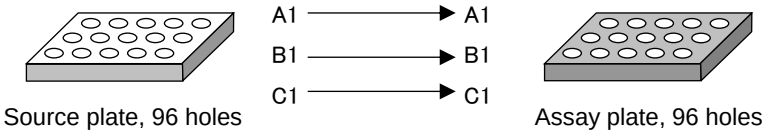
After all compound has been dripped onto the assay plate, the used tip is moved to the tip disposal location and is disposed of as a waste.

Dispensing Pattern

Since the number of wells in the source plate does not always match the number of wells in the assay plate, the following examples of combinations are to be considered. Only one source plate can be set, while up to four assay plates can be set.

1) 96-hole Source plate → 96-hole Assay plate

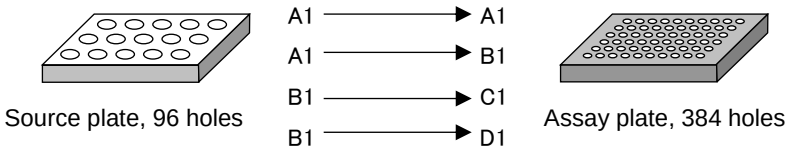
This is the simplest pattern where the wells in one plate have a one-to-one association with the wells in the other plate.



Source plate, 96 holes　　Assay plate, 96 holes

2) 96-hole Source plate → 384-hole Assay plate

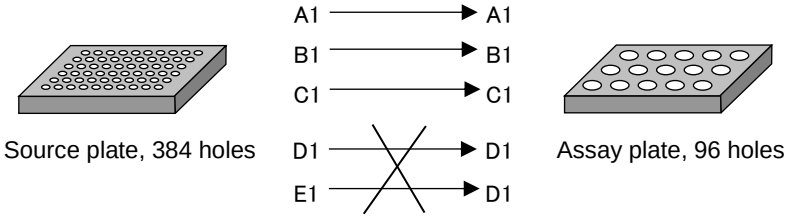
In this combination, one well in the source plate is associated with multiple wells in the assay plate.



Source plate, 96 holes　　Assay plate, 384 holes

3) 384-hole Source plate → 96-hole Assay plate

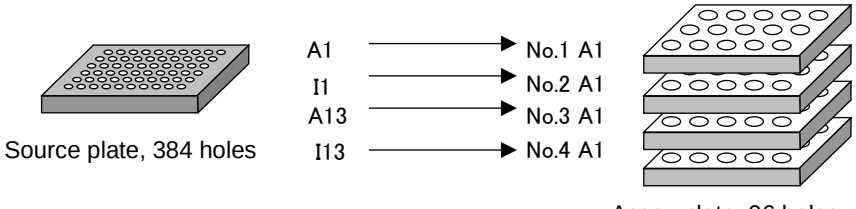
Dispensing from the multiple wells in the source plate to one well in the assay plate cannot be performed.



Source plate, 384 holes　　Assay plate, 96 holes

4) 384-hole Source plate → 96-hole Assay plate × 4

In this combination, one well in the source plate is associated with one well in one of the assay plates. This dispensing pattern is selectable when a large incubator (sold separately) is used.



Source plate, 384 holes　　Assay plate, 96 holes

Dispensing Actions

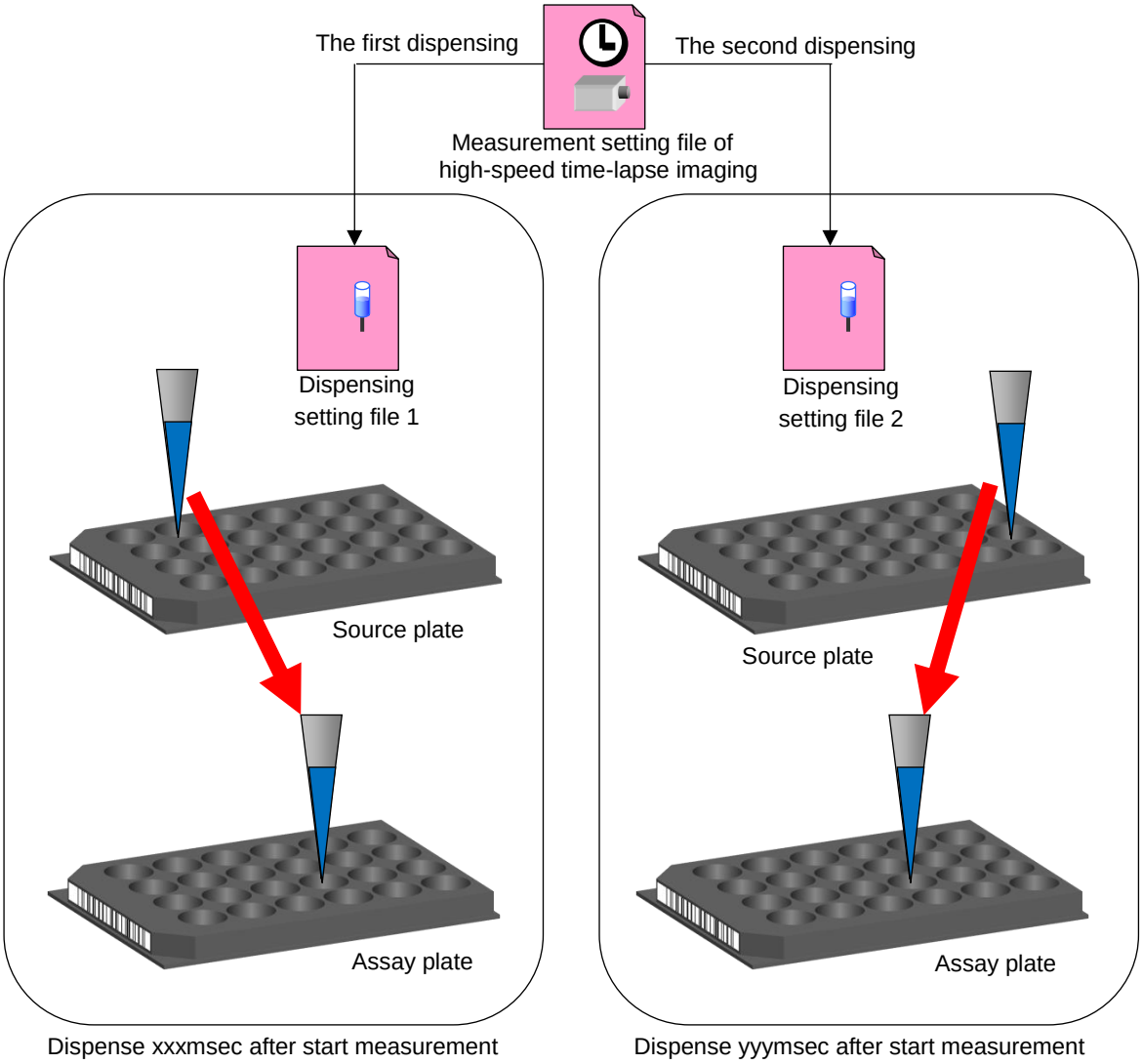
Ten steps of dispensing actions, including filling, dripping and agitation, can be set for each dispensing operation performed between source and assay plates. Detailed parameters such as the dripping speed and amount, and whether or not to implement agitation, can be set for each dispensing action.

List of dispensing actions

Dispensing action	Explanation
Liquid Surface	The liquid surface is detected. The tip is lowered within the well in the source plate until its edge contacts the liquid, to detect the position of liquid surface in the well.
Prewet	Solution is filled and then dripped within the well in the source plate to wet the inside of the tip with the solution.
AspirateStir	Solution is agitated within the well in the source plate. Solution is suctioned and discharged for the specified number of times.
Airgap	Air is introduced into the tip before solution. This way, all solution can be dripped.
Aspirate	Solution is filled from the well in the source plate.
AspirateTiptouch	The tip is caused to contact the wall of the well to remove any water droplet attached at the edge of the tip.
Dispense	Reagent is dropped onto the assay plate.
DispenseStir	Dripped reagent is agitated in the well. Solution is suctioned and then discharged within the well for the specified number of times.
DispenseTiptouch	The tip is caused to contact the wall of the well to remove any water droplet attached at the edge of the tip.
Air Blow	This allows to blow air to drop droplet adherent on the tip top.

Multi Dispensing

In high-speed time-lapse imaging, it is possible to dispense from wells from source plate to one well of assay plate. Disposable tips are exchanged after each dispensing is performed. Before you do multi dispensing, you make setting files of each dispensing. And when you start measurement, you assign these setting files and dispensing is done as the order which you assign them.



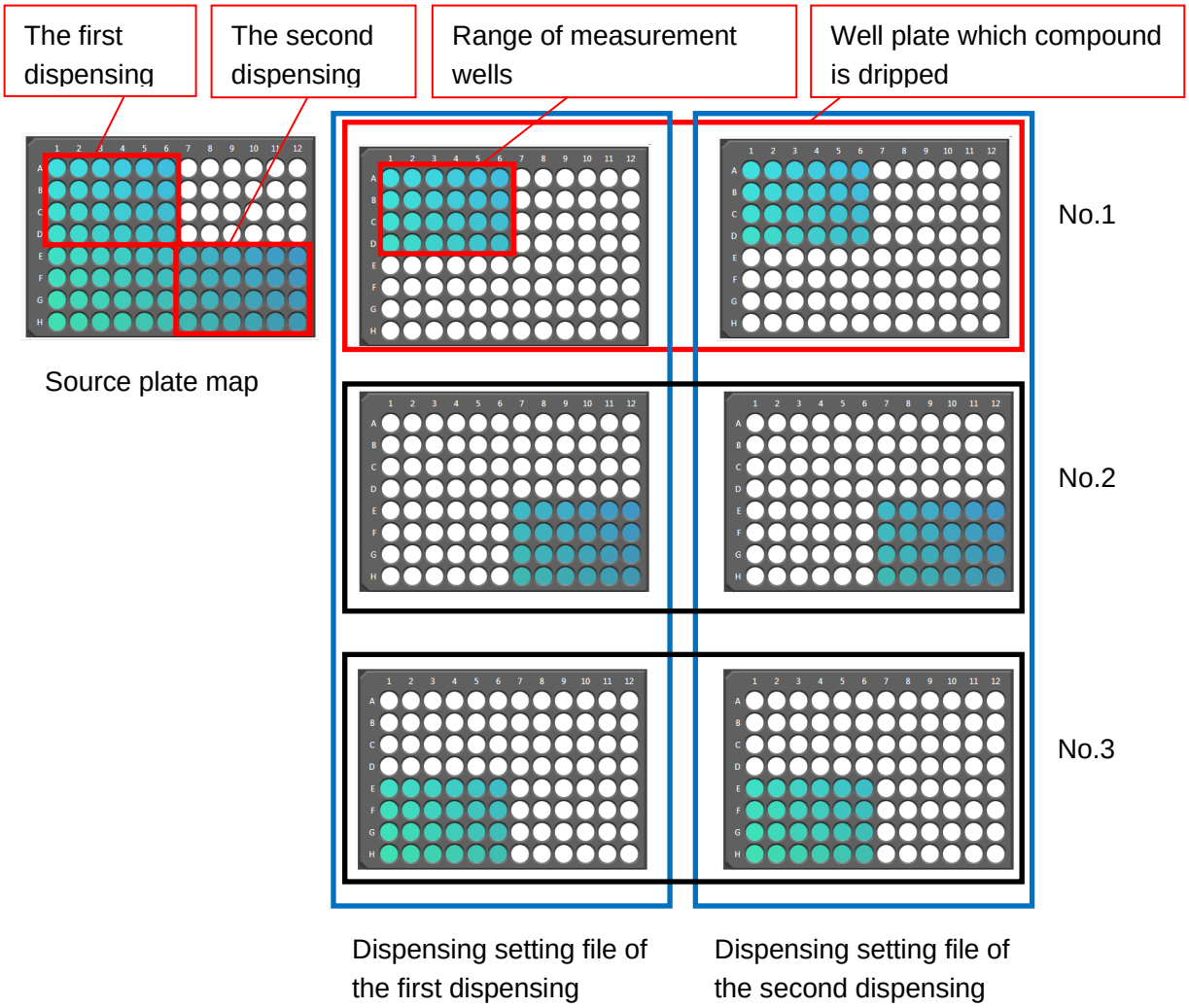
Working of imaging with multi dispensing

In high speed time lapse imaging, wells for imaging is decided as setting of imaging region in measurement setting file. Settings of multi dispensing need to meet following condition.

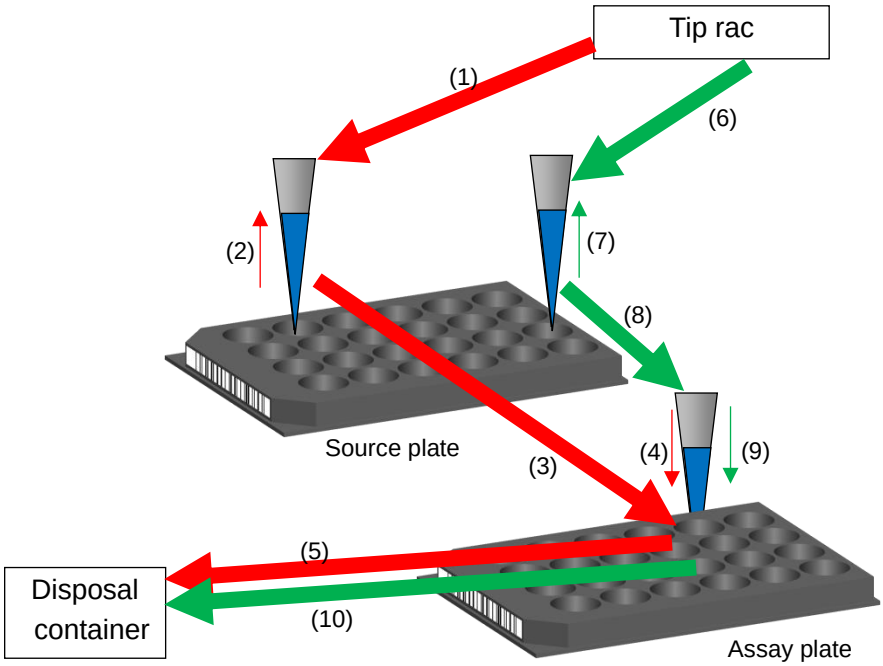
- Make dispensing setting file of each dispense.
  (For example, if you perform three times of dispensing, three dispensing setting files is needed)
- Well numbers of well-plate map of source plates must be set as same in all dispensing setting files.
- Well numbers of all well-plate maps of assay plates must be set as same in

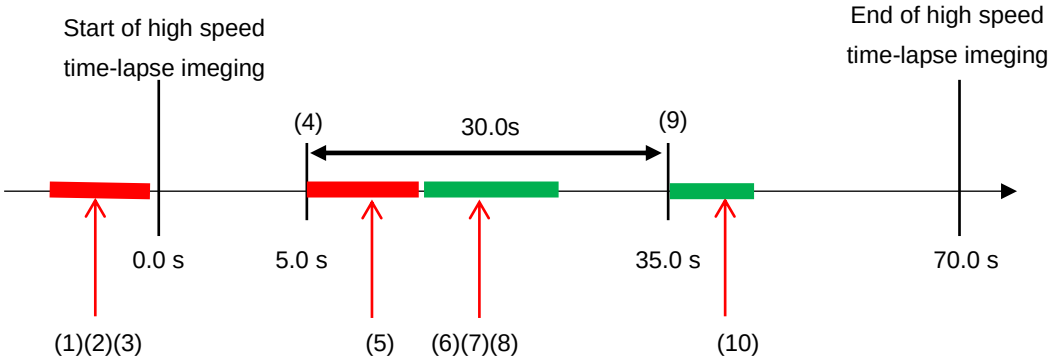
all dispensing setting files.

- ●Wells which compound is dripped of each well-plate map number of assay plate must be set as same in all dispensing setting files.
- ●Number of well-plate map of assay plate must be set as same in all dispensing setting files.
- ●Measurement wells assigned in measurement setting file must be selected from the well-plate map of dispensing setting file.
- ●Only one well plate which compound is dripped can be assigned from well-plate map (No.1 – No.4) of assay plate which is made in dispensing setting file. It is impossible to dispense to multiple assay plates.
- ●In case of multi dispensing with robot handling system, No. 1 well-plate map is applied as well-plate which compound is dripped from well-plate map of assay plate which is made in dispensing setting file.
- ●30 seconds of intervals are needed between procedures of multi dispensing



Example of multi dispensing setting





Exapmle of measurement with multi dispensing

(1) Take out a tip from tip rack
(2) Suction the compound from source plate
(3) Move to assay plate
「Start high-speed time-lapse imaging」
(4) Drip the compound to assay plate 5 second after starting of high-speed time-lapse imaging
(5) Discard the tip
(6) Take out the new tip from tip rack
(7) Suction the other compound from source plate
(8) Move to assay plate
(9) Drip the compound to assay plate 35 second after starting of high-speed time-lapse imaging
(10) Discard the tip
「70 second after start imaging, stop high-speed time-lapse imaging」

Imaging channels

On the Image Setting screen, imaging channels are set. A combination of object lens, light source, filter wheel and camera settings is stored for each imaging channel.

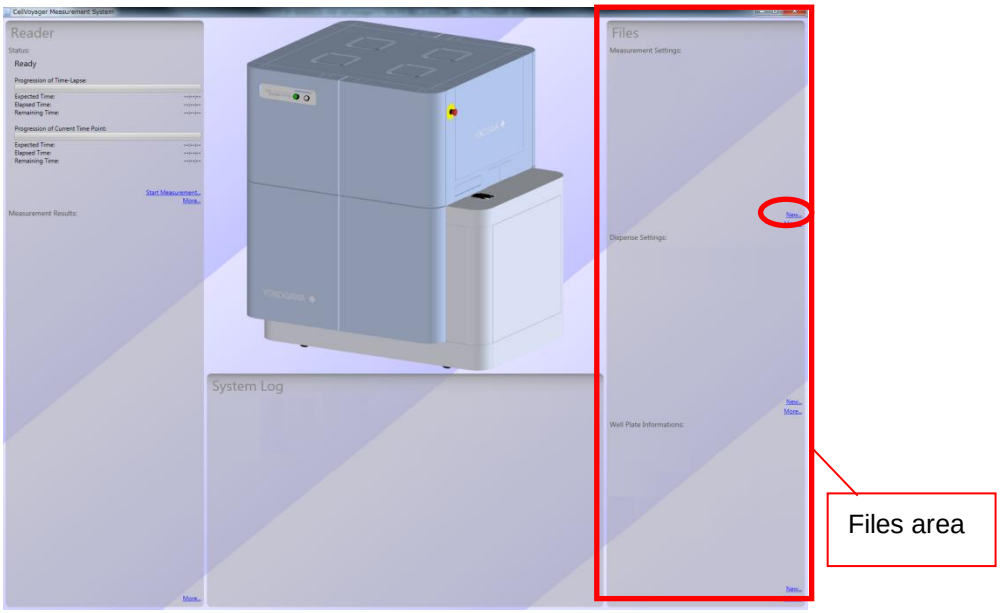
List of setting items for each imaging channel

Item		Explanation
Ch		Number to identify the imaging channel
Target		Specify the name of the target to be imaged.
Method		Select the combination of filter and light source.
Objective		Select the object lens.
Acquisition		Select the filter name to be used for imaging. A filter name list is generated from the filters selected on the filter wheel.
ExposureTime		Specification of exposure time. Unit：ms
Binning		Select the binning. 1x1, 2x2, 3x3 or 4x4.
LightSource		Select the laser as the light source. If the laser is selected, multiple wavelengths can be combined.
Fluorophore		Select the used fluorophre. (It is needed for performing crosstalk correction by Image Correction Software)
Preview	Color	Specify the display color for preview in the #rrggbb format. Default color is subject to filter setting selected by “Acquisition.”
	MinLevel	Specify the lower limit of input signals for generating a preview in a range of 0.0 to 1.0.
	MaxLevel	Specify the upper limit of input signals for generating a preview in a range of 0.0 to 1.0.

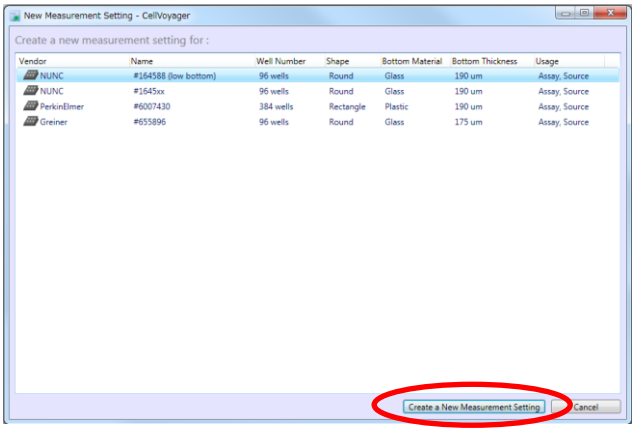
5.2. Displaying the Measurement setting File Screen

Creating a New Measurement Setting File

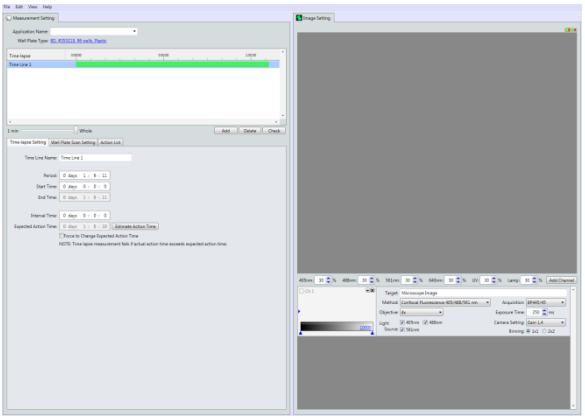
1) Click “New” in the Files area.



2) Select a desired well plate product and click “Create a New Measurement Setting.”



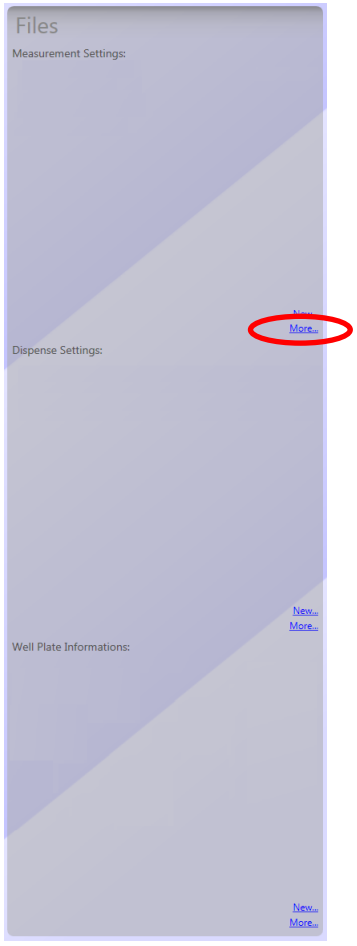
The screen for editing a measurement setting files opens. (Refer to 6.2)



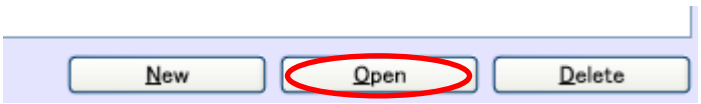
* If the plate you wish to use is not on the list, register the plate by referring to 5.16.

Editing a Measurement Setting File

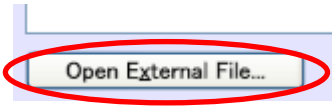
1) Click “More” at the bottom of the Measurement Settings section in the Files area.



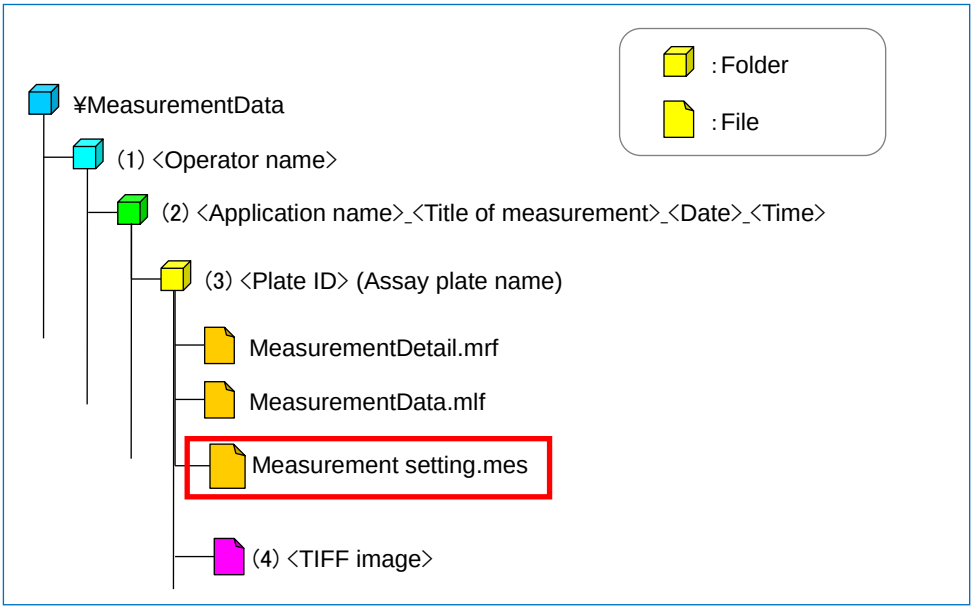
2) Select a desired measurement setting file and click “Open.”



Click “Open External File” if you open the measurement setting file from the external folder you saved.



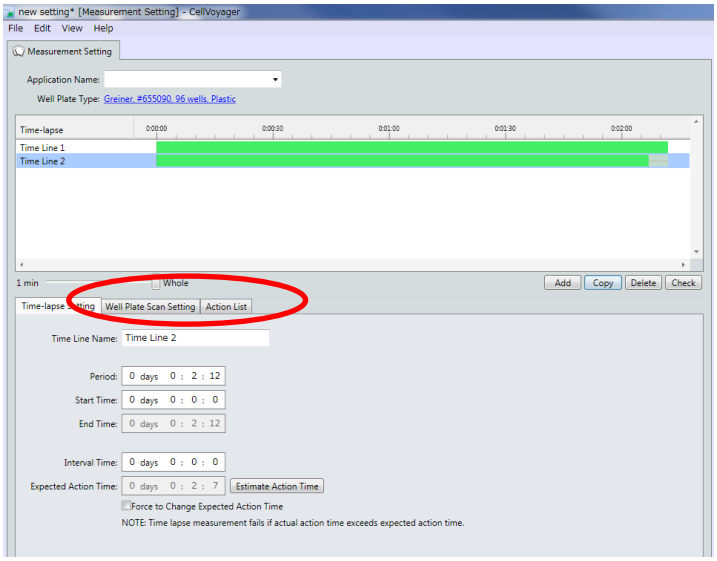
Select the file whose extension name of “.mes”. The measurement setting file is shown.



5.3. Setting a Time Line

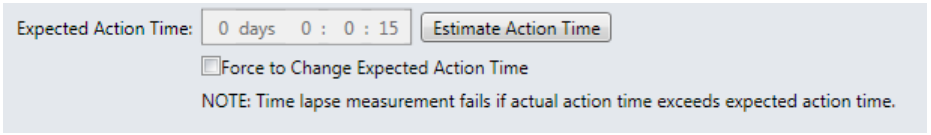
Each measurement setting file retains various setting information for measurement processing along a time line. For each time line, set the well movement pattern, imaging points and measurement actions such as fluorescence imaging and 3D imaging.

1) Set the items on the Well Plate Scan Setting tab（refer to 5.6) and Action List tab(refer to 5.7).

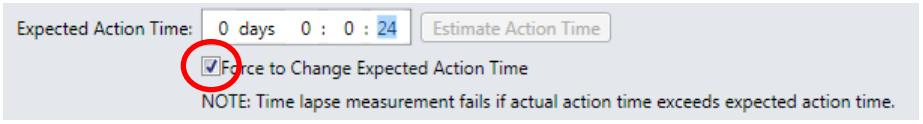


2) Click the Time-lapse Setting tab. (Refer to 6.2)

3) Click “Estimate Action Time” to display the expected time of measurement.



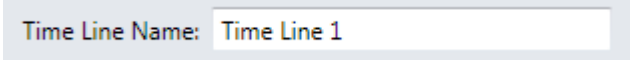
The Expected Action Time field can be entered if the Force to Change Expected Action Time checkbox is checked.



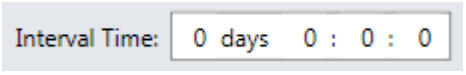
MEMO

●The time lapse measurement fails if the actual action time exceeds the expected action time.

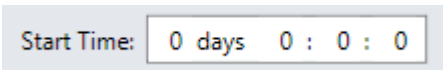
4) Enter the name of the time line. (You can use the default name.)



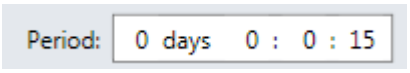
5) To perform time-lapse imaging (refer to 5.1) for each well plate, enter the imaging interval. Enter a value greater than the one shown in the Expected Action Time field.



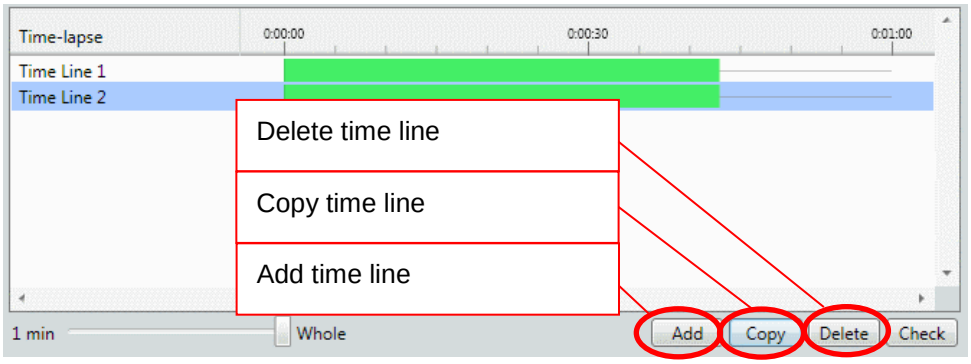
6) Enter the start time of time line.



7) Enter the end time of the time line.



8) Click "Add," and a new time line will be created. Select Time Line 2 and set the items on the Well Plate Scan Setting tab and Action List tab, as well as time-line timings, for Time Line 2.
(Click “Copy” to copy time line. Click “Delete” to delete.)



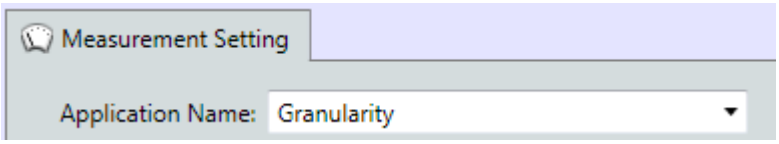
MEMO

●Click “Copy” button, and same time line as selected is copied to the last of list.
In case that dispensing setting is included in selected time line, it is impossible to “Copy”.

9) Click “Check,” and overlaps of time lines will be adjusted automatically. You can extend or shorten each time line using the scale bar.

5.4. Entering the Application Name

Enter the application name such as “Granularity”.
A desired name can be entered. (The field may remain blank.)



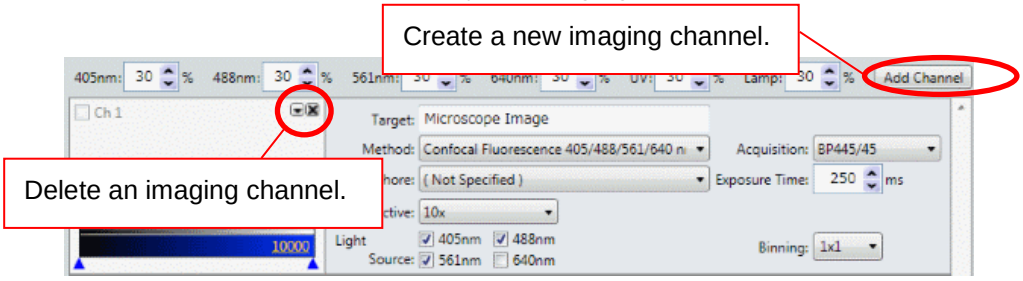
5.5. Setting the Imaging Channel

Set the object lens, light source, filter, CSU and sCMOS camera.

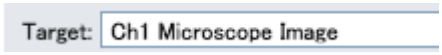
MEMO

●Image channel settings may be different by specifications.
Please confirm your specification items for CV7000.

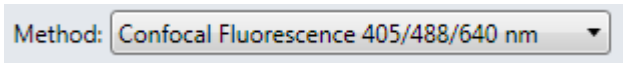
1) Click “Add Channel” and specify the imaging channel number. (Refer to 6.2.)



2) Enter the name of the imaging target.



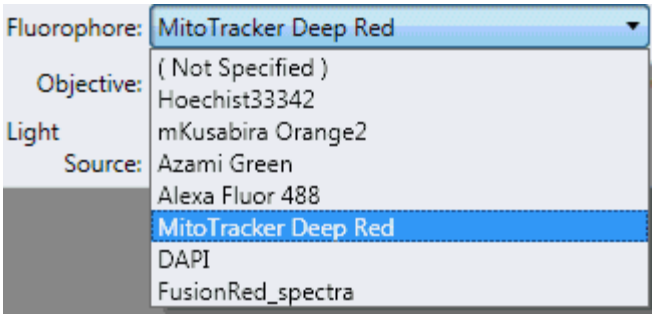
3) Select the combination of optical systems used for imaging.



Optical system	Explanation
Confocal Fluorescence 405/488/561(or 532)nm	Confocal imaging using a light source of 405, 488 or 561(or 532) nm
Confocal Fluorescence 405/488/640nm	Confocal imaging using a light source of 405, 488 or 640 nm
Confocal Fluorescence 405/488/561/640nm (QUAD-DM model only)	Confocal imaging using a light source of 405, 488, 561 or 640nm
Epifluorescence 405/488/561(or 532)nm	Epifluorescence imaging using a light source of 405, 488 or 561(or 532) nm
Epifluorescence 405/488/640nm	Epifluorescence imaging using a light source of 405, 488 or 640 nm
Epifluorescence 405/488/561/640nm (QUAD-DM model only)	Epifluorescence imaging using a light source of 405, 488, 561 or 640nm
Epifluorescence UV Lamp (UV lamp model only)	UV light source

Brightfield (Bright field model only)	Brightfield imaging to match pixel position with epifluorescent imaging
Brightfield(Confocal path) (Bright field model only)	Brightfield imaging to match pixel position with confocal imaging
Phase Contrast (Phase contrast model only)	Phase contrast imaging to match pixel position with epifluorescent imaging.
Phase Contrast(Confocal path) (Phase contrast model only)	Phase contrast imaging to match pixel position with confocal imaging
Digital Phase Contrast (Bright field model only)	DPC imaging to match pixel position with epifluorescent imaging.

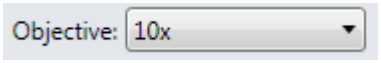
4) Select the fluorophore



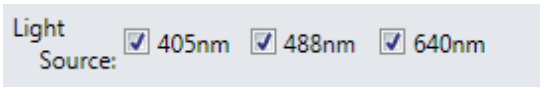
MEMO

- If crosstalk correction will be performed by Image Correction Software, fluorephore must be selected,
- Refer to 5.15 about registration of fluorophore.

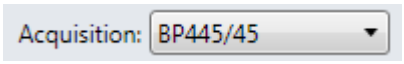
5) Select the object lens.



6) Select the light source used for imaging.

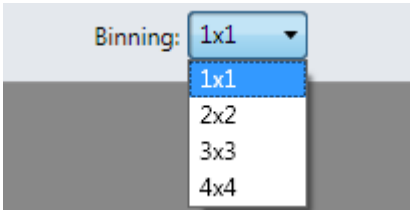


7) Select the fluorescence filter.



8) Set the exposure time. (11ms≦Exposure Time≦9999ms)

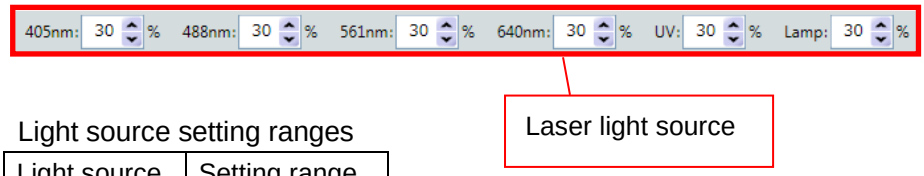
9) Set camera binning.



MEMO

●If multiple channels have been set, it is recommended to use the same binning setting for all channels. However CV7000 Analysis Software supports different binning setting on multiple channels, errors in recognition result can occur.

10) Set the light source output.

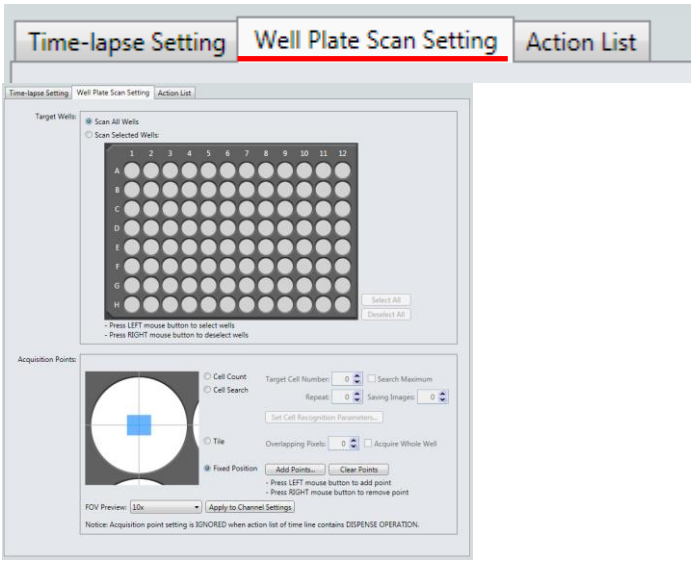


Light source setting ranges

Light source	Setting range
405nm	0, 10 to 100%
488nm	0, 10 to 100%
561nm	0, 10 to 100%
640nm	0, 10 to 100%
UV	0 to 100%
Lamp	0 to 100%

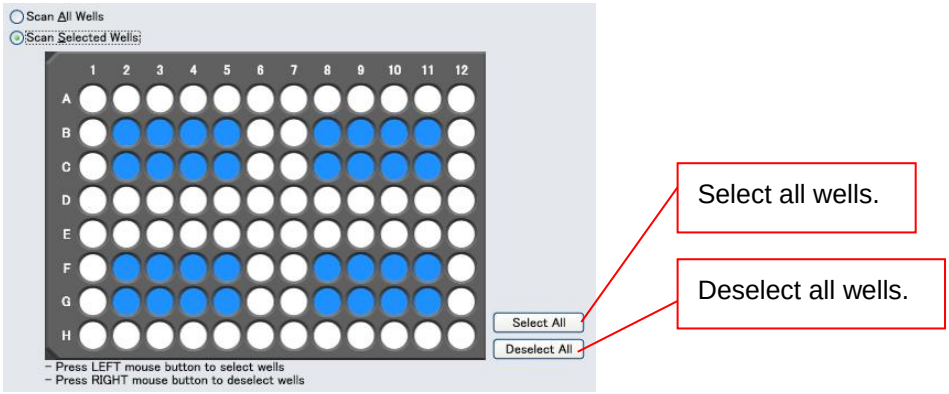
5.6. Setting Acquisition Points in the Well

1) Set the well acquisition points. Click the Well Plate Scan Setting tab.

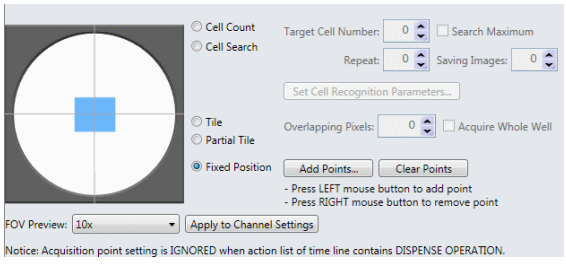


2) Select the wells to scan.

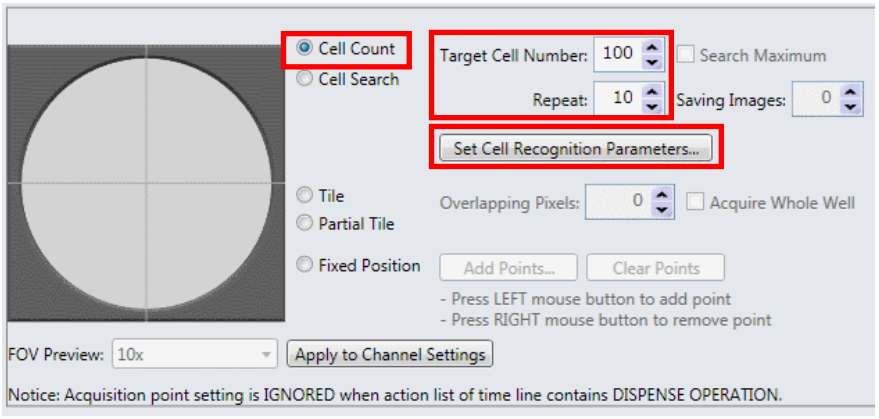
When you select Scan Selected Wells, select the wells to scan.
Left-click to select a well.
Right-click to deselect a well.



3) Set acquisition points in the well. Acquisition points can be set in one of four modes including “Cell Count,” “Cell Search,” “Tile”, “Partial Tile” and “Fixed Position.” (Refer to 5.1)



Cell Count Function

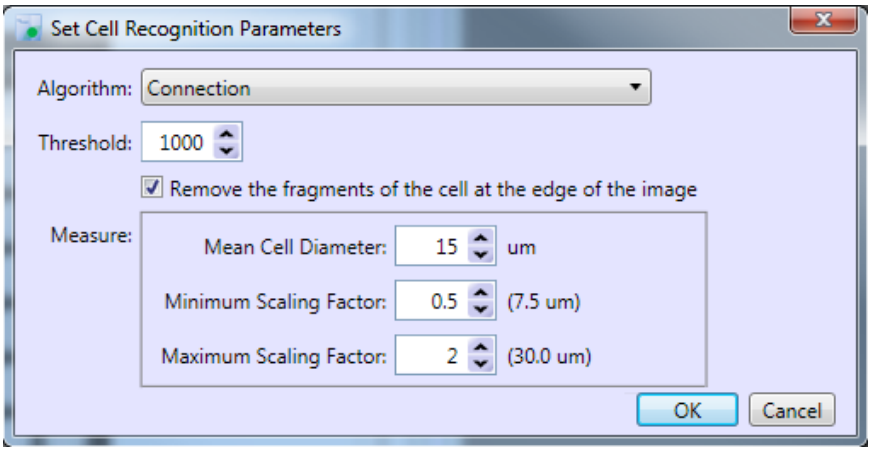


1) Select “Cell Count.” (Refer to 5.1 and 6.2)

2) Enter the value of “Target Cell Number” (specified cell count).

3) Enter the value of “Repeat” (maximum number of images to be captured).

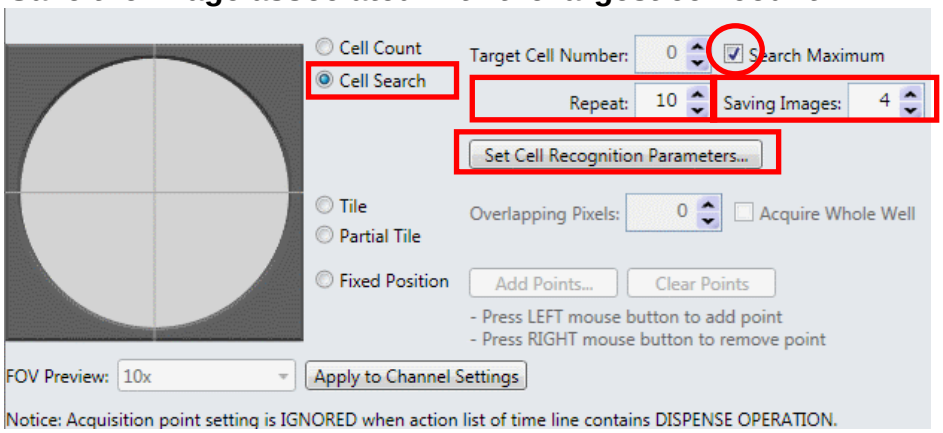
4) Click “Set Cell Recognition Parameters.” The screen for setting the cell recognition algorithm opens.



5) Set the cell recognition algorithm. (Refer to 5.10)

Cell Search Function

Save the image associated with the largest cell count



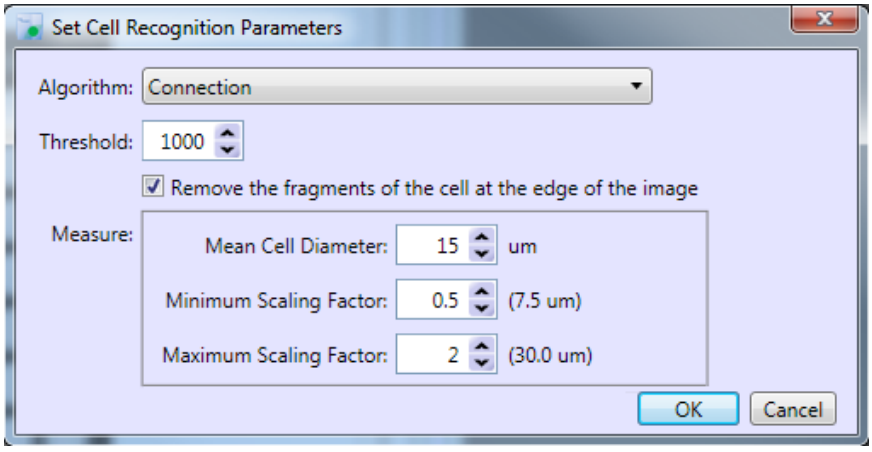
1) Select “Cell Search.” (Refer to 5.1 and 6.2)

2) Select “Search Maximum.”

3) Enter the value of “Repeat” (number of images to be captured)

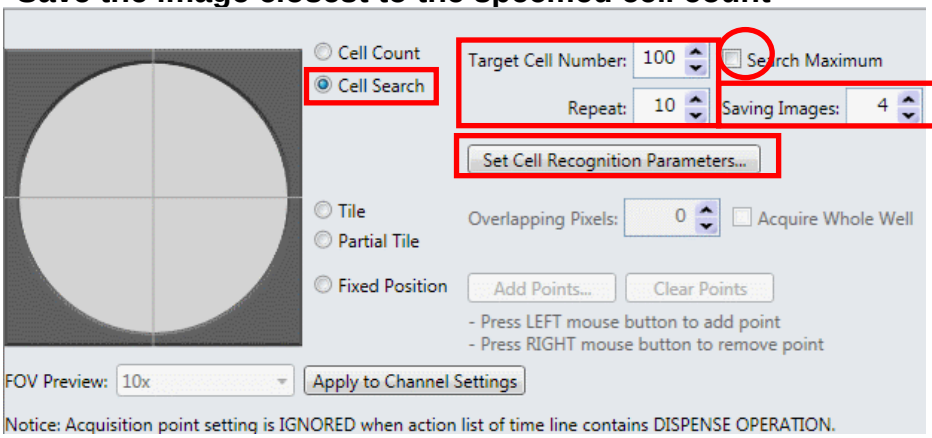
4) Specify the value of “Saving Images” (number of images to be saved).

5) Click “Set Cell Recognition Parameters.” The screen for setting the cell recognition algorithm opens.



6) Set the cell recognition algorithm. (Refer to 5.10)

Save the image closest to the specified cell count



1) Select “Cell Search.” (Refer to 5.1 and 6.2)

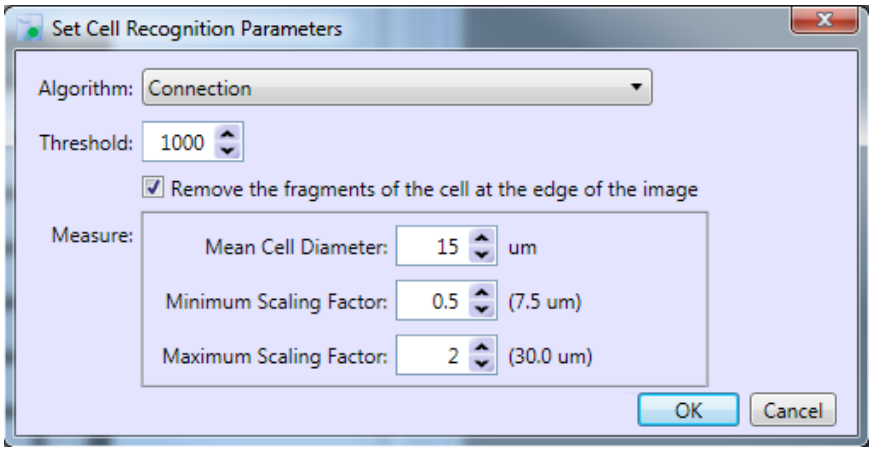
2) Unselect the “Search Maximum” check box.

3) Enter the value of “Target Cell Number” (specification of cell count).

4) Enter the value of “Repeat” (number of images to be captured).

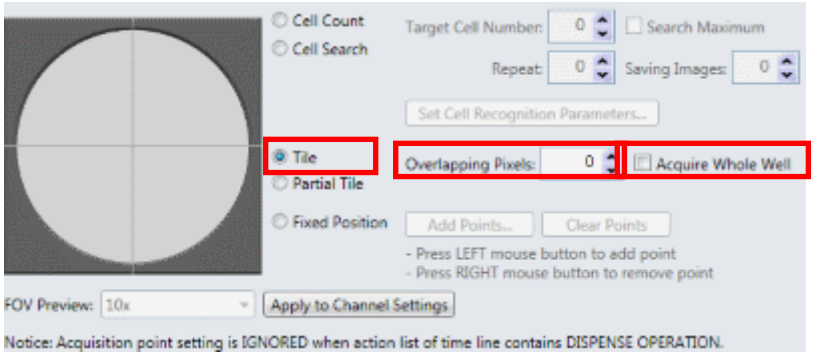
5) Specify the value of “Saving Images” (number of images to be saved). Images will be saved one by one, starting from the image closest to the specified cell count.

6) Click “Set Cell Recognition Parameters.” The screen for setting the cell recognition algorithm opens.



7) Set the cell recognition algorithm. (Refer to 5.10)

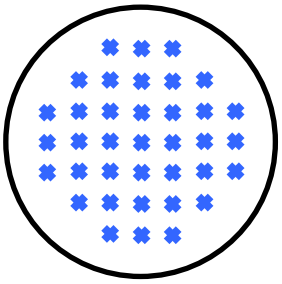
Tile Function



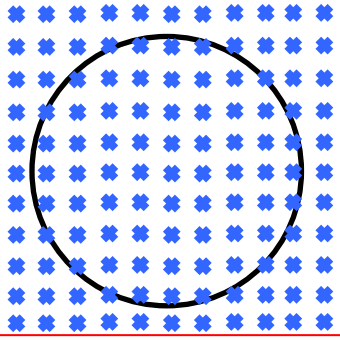
1) Select “Tile.” (Refer to 5.1 and 6.2)

2) Enter the value of “Overlapping Pixels” (number of overlapping pixels of images). To have images overlap with each other, enter a positive value. To keep images apart, enter a negative value. (-100≦Overlapping Pixels≦100) Recommended value of Overlapping Pixels is “50”.

3) The fields of whole well are acquired if “Acquire Whole Well” is checked.



An example that “Acquire Whole Well” is not checked.



An example that “Acquire Whole Well” is checked.

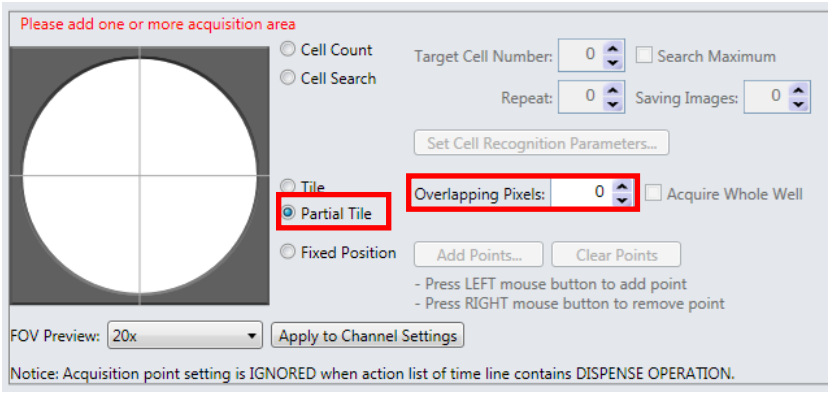
Reference numbers of images captured by the tile function excluding edge areas (per well)

Magnification	96 wells	384 wells
10x	6 images	-
20x	36 images	6 images
40x	194 images	48 images
60x	470 images	130 images

Reference numbers of images captured by the tile function including edge areas (per well)

Magnification	96 wells	384 wells
10x	20 images	6 images
20x	80 images	20 images
40x	304 images	80 images
60x	696 images	168 images

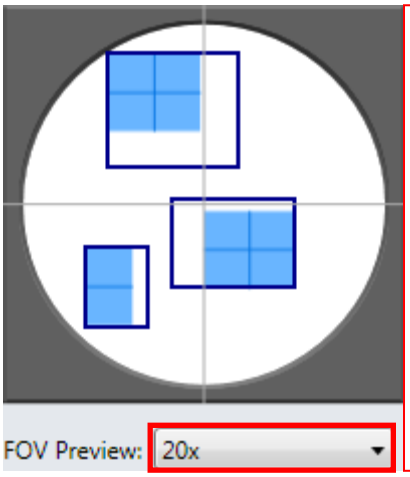
Partial Tile Function



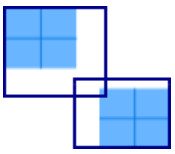
1) Select “Partial Tile.” (Refer to 5.1 and 6.2)

2) Enter the value of “Overlapping Pixels” (number of overlapping pixels of images). To have images overlap with each other, enter a positive value. To keep images apart, enter a negative value. (-100≦Overlapping Pixels≦100) Recommended value of Overlapping Pixels is “50”.

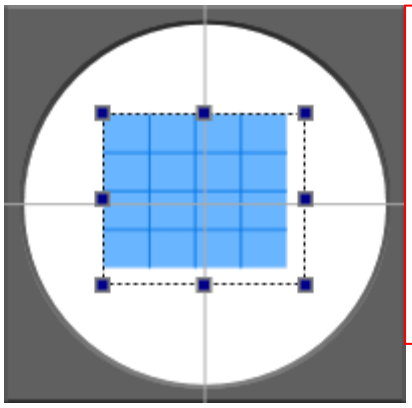
3) Designate acquisition region
You can left-drag the well to specify desired acquisition region. Right-drag the selected region to delete the region.



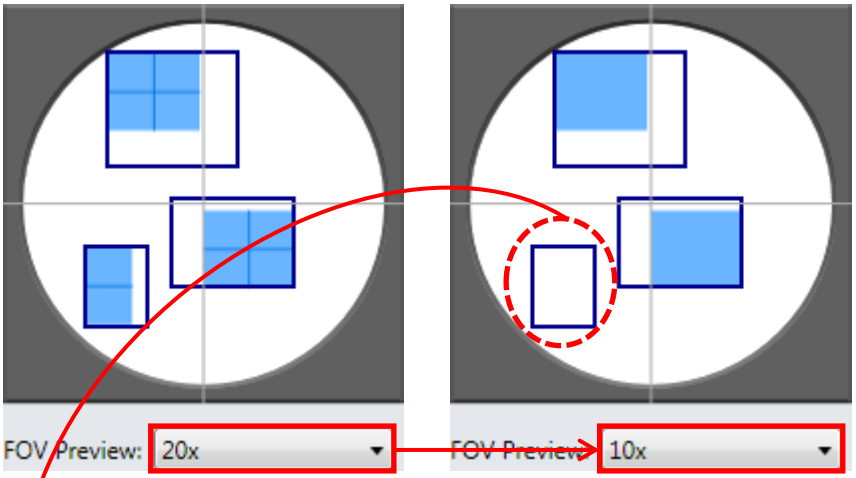
Select objective lens to show the preview and select acquisition region by left-dragging
Plural regions can be designated
Each region can’t be overlapped

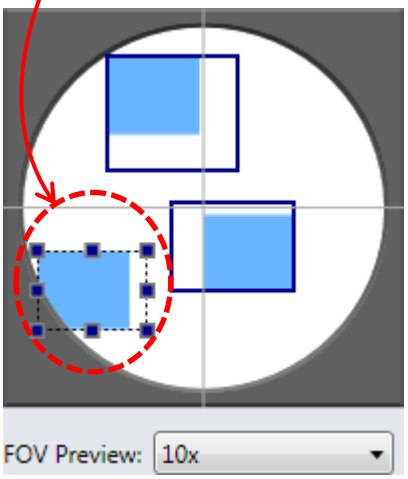


NG

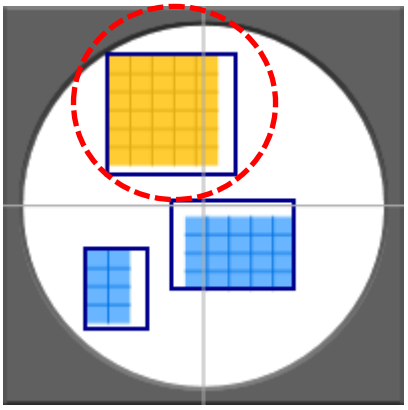


Display of selected region become as left diagram by left-clicking on the region
The region can be moved by left-dragging
The region can be expanded/ reduced by left-dragging 4 sides/ 4 corners of region





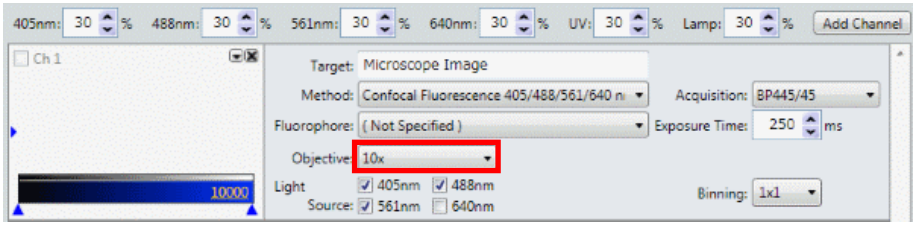
If objective lens for preview is changed to lower magnification after designating regions, the regions can be smaller than 1 Field
In the case, please change to higher magnification lens or expand the regions



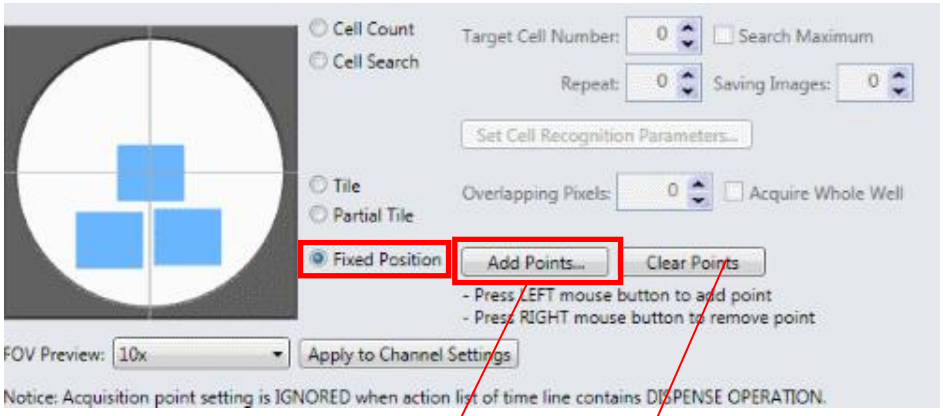
The indication of maximum Fields number that a tiled image can include to analyze by CV7000 Analysis Software is 25 Fields (sCMOS model)
If the region is designated so that over the Fields, they become orange
(No problem in imaging)

The restriction in imaging region can exist in some sample vessels when water immersion objective lens is used. In this case, prohibited area is displayed as pink. (refer to right of above diagram)
Prohibited area s are unselectable as imaging region. However, if objective lens for preview is changed to water immersion type from dry type after designating regions, the regions can be in prohibited area. In the case, please reset the imaging regions.

Objective lens selected in above procedure is to preview for setting region of tiling. Select actual objective lens used for measurement in channel setting window. (refer to 5.5).

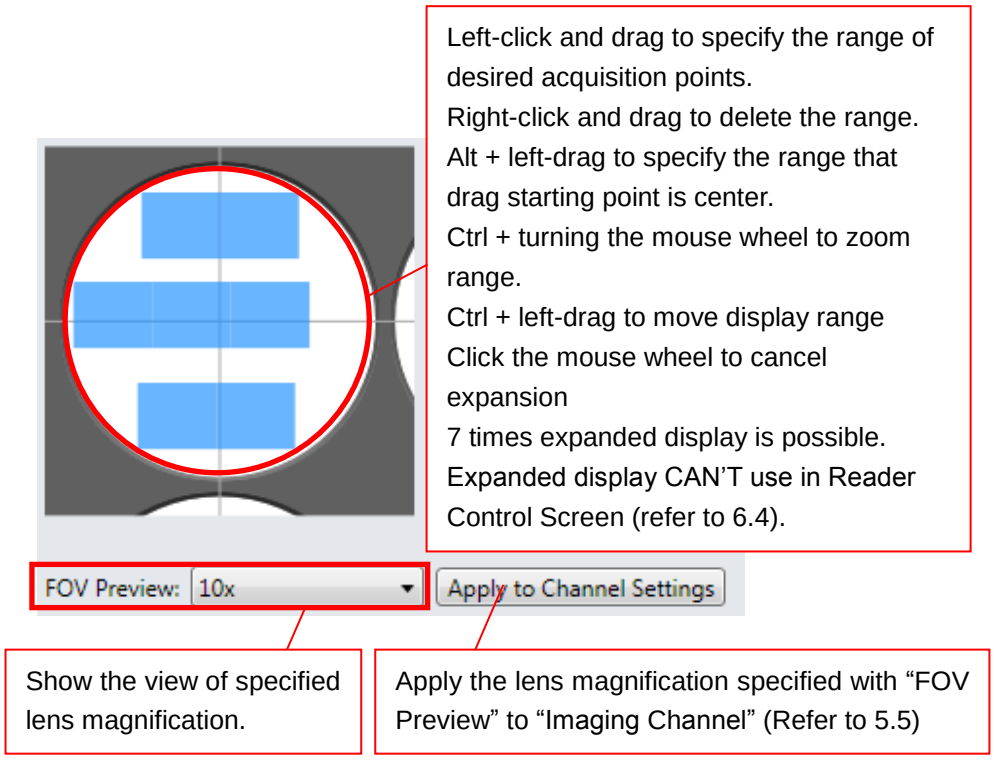


Fixed Position Function

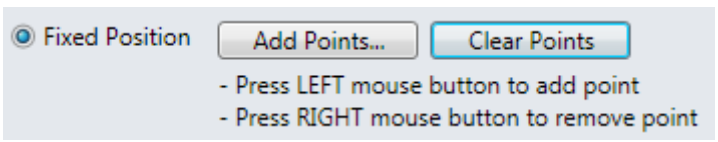


1) Select “Fixed Position.” (Refer to 5.1 and 6.2)

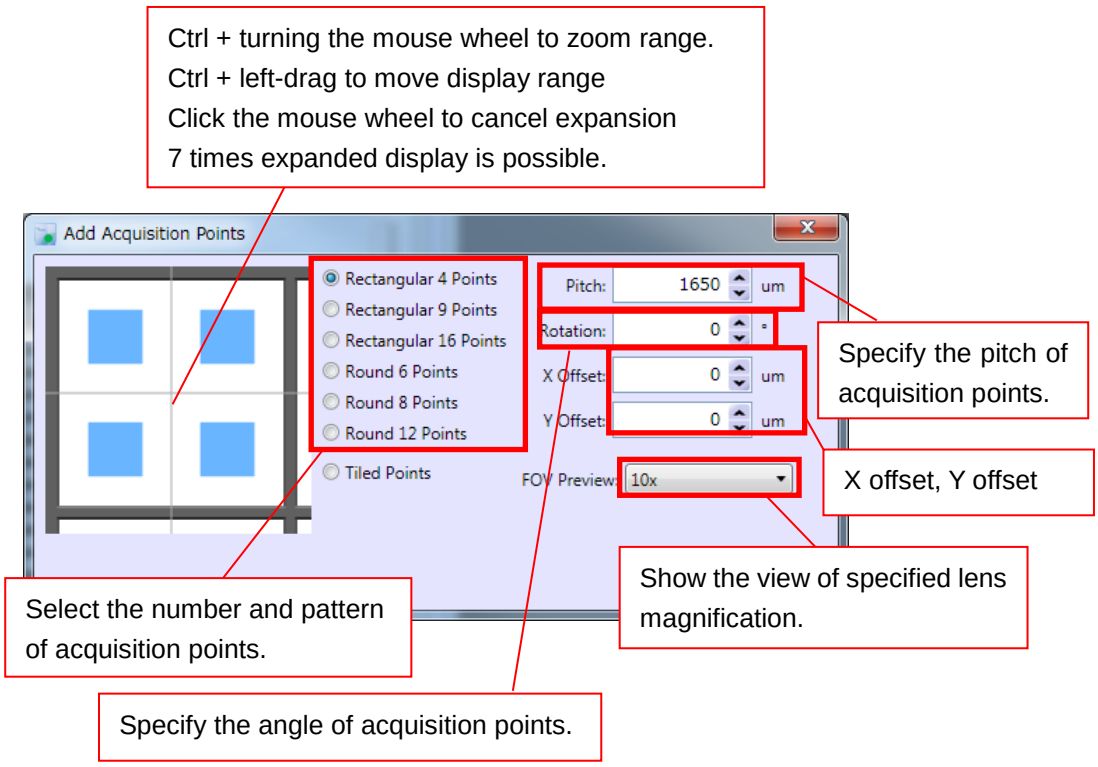
2) Specify the acquisition points.
You can left-click the well to specify desired acquisition points. Right-click an acquisition point to delete the point.



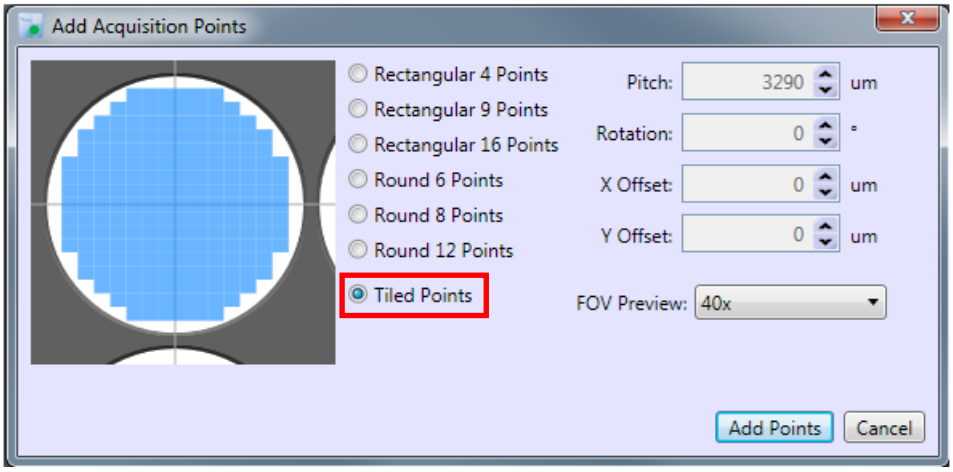
To specify a pattern, which is a combination of number of acquisition points, interval, etc., click “Add Points.”



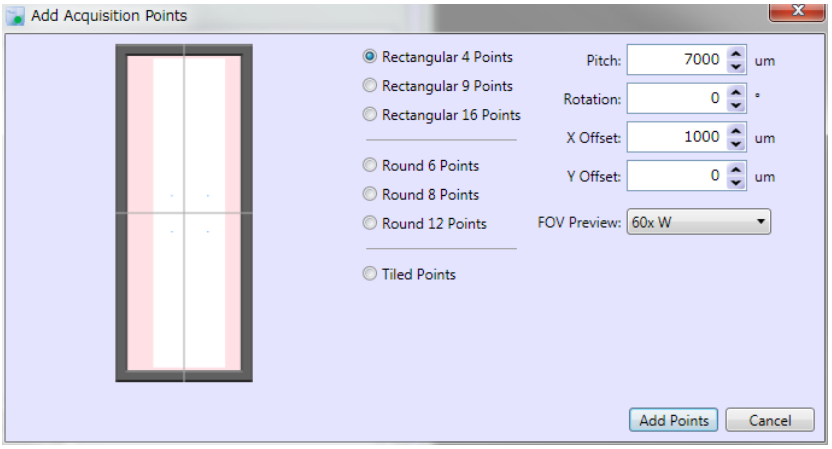
3) After clicking “Add Points,” set the number, pattern, pitch and angle of imaging points. After all items have been set, click "Add Points” again.



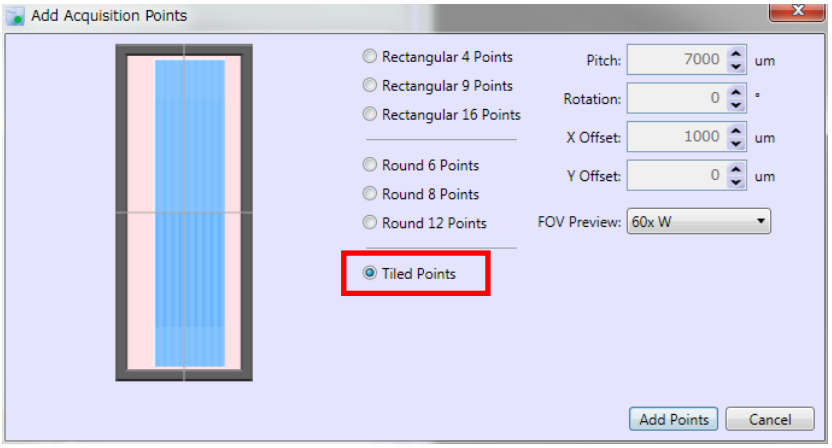
If selecting “Tiled Points,” set as acquisition points the view field to be captured at a specified magnification factor in a tiled pattern. (Acquisition points can be added or deleted after clicking “Add Points.”)



The restriction in imaging region can exist in some sample vessels when water immersion objective lens is used. In this case, prohibited area is displayed as pink. (refer to right of above diagram)
Prohibited area is unselectable as imaging region. However, if objective lens for preview is changed to water immersion type from dry type after designating regions, the regions can be included in prohibited area. In the case, please reset the imaging regions.



In case that select “Tiled Points” when imaging prohibited region exists, tiled region is selected inside acquirable region as below diagram.



MEMO

●Difference between the Tile Function and “Tiled Points”

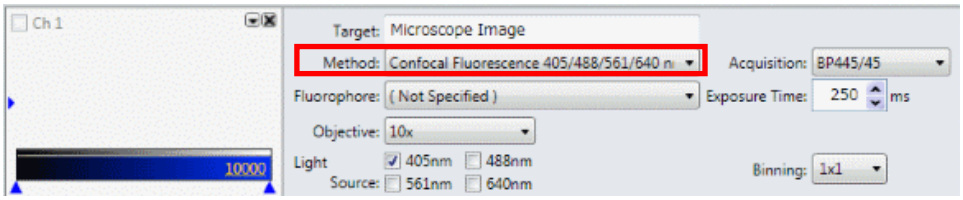
The tile function lets you capture the entire view field of the well in a tiled pattern using the object lens selected when the imaging channel was set (refer to 5.5). You can also specify the number of pixels to overlap between tiled images.
With “Tiled Points,” specify as acquisition points the view field to be captured at a specified lens magnification factor in a tiled pattern. You can also add or delete acquisition points.

5.7. Settings on the Action List Tab

Software Focus Setting

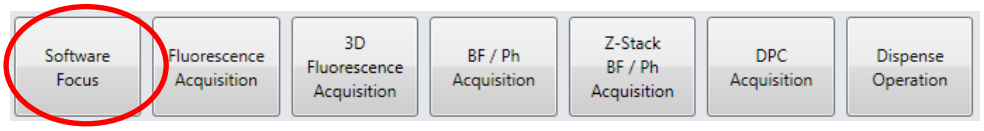
Refer to 5.1 for the software focus function.

1) Set the imaging channel. Under “Method,” select “Confocal Fluorescence.” (Refer to 5.5)

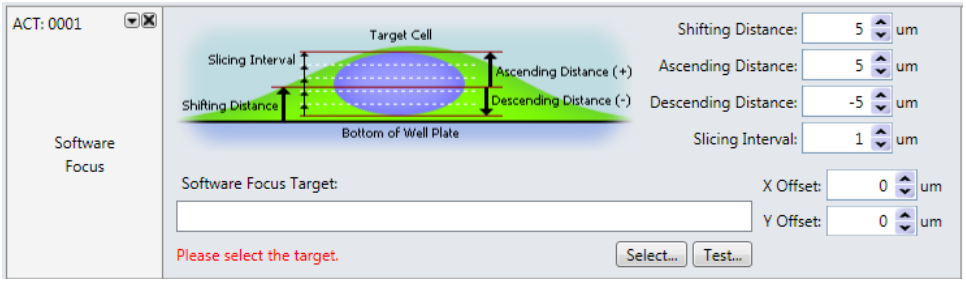


2) Click the Action List tab.

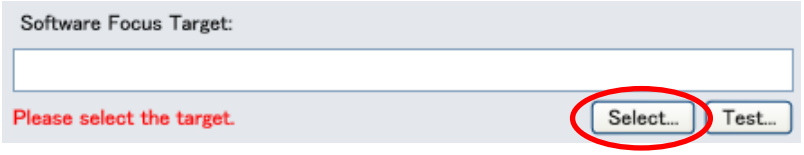
3) Click “Software Focus.”



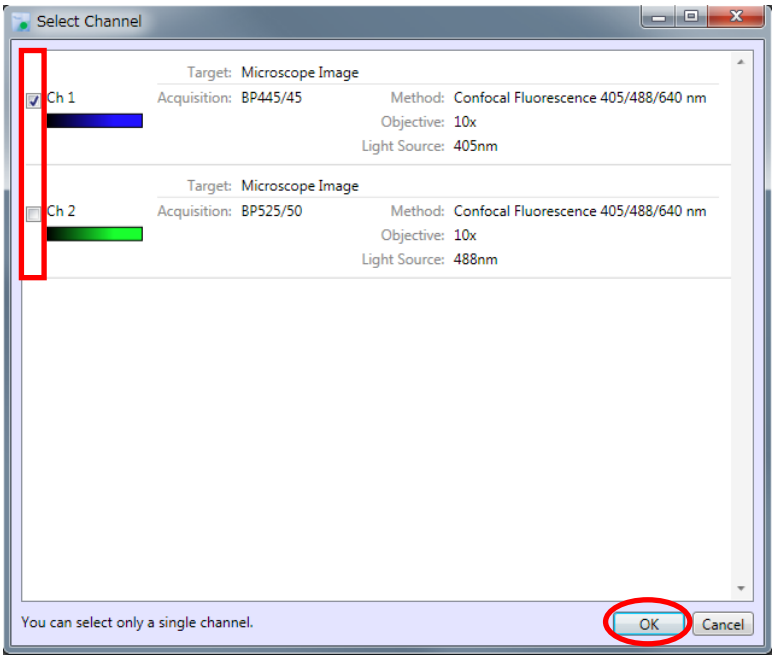
The screen for setting the software focus opens. (Refer to 6.2)



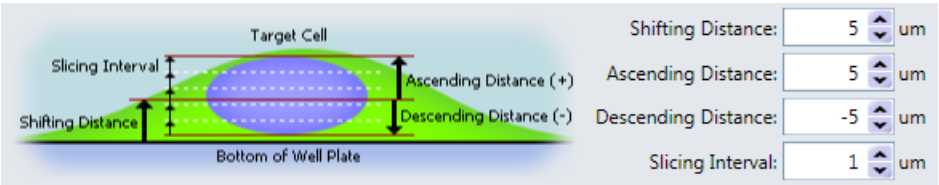
4) Click “Select.”



5) The Select Channel screen opens. Select the target channel, and then click "OK."



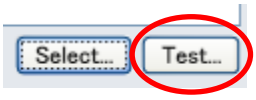
6) Set the imaging area based on software focus.



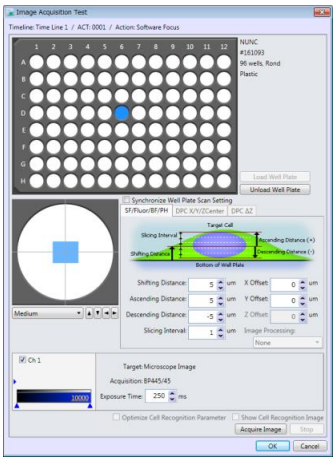
Item	Explanation
Shifting Distance	Amount of shift from the auto-focus position Reference plane of software focus
Ascending Distance	Distance from the Shifting Distance position to the top plane of software focus
Descending Distance	Distance from the Shifting Distance position to the bottom plane of software focus
Slicing Interval	Z step width

In the above example, the plane 5 µm above the auto-focus position is set as the reference plane of software focus, and images are captured in 1 µm steps over the area between 5 µm below and 5 µm above this focal plane. From the total of 11 images captured, the one with the highest average brightness is output as the image in focus.

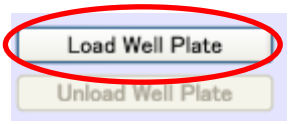
7) Display an imaging preview to check the imaging settings. Click "Test."



The Image Acquisition Test screen opens. (Refer to 6.2)



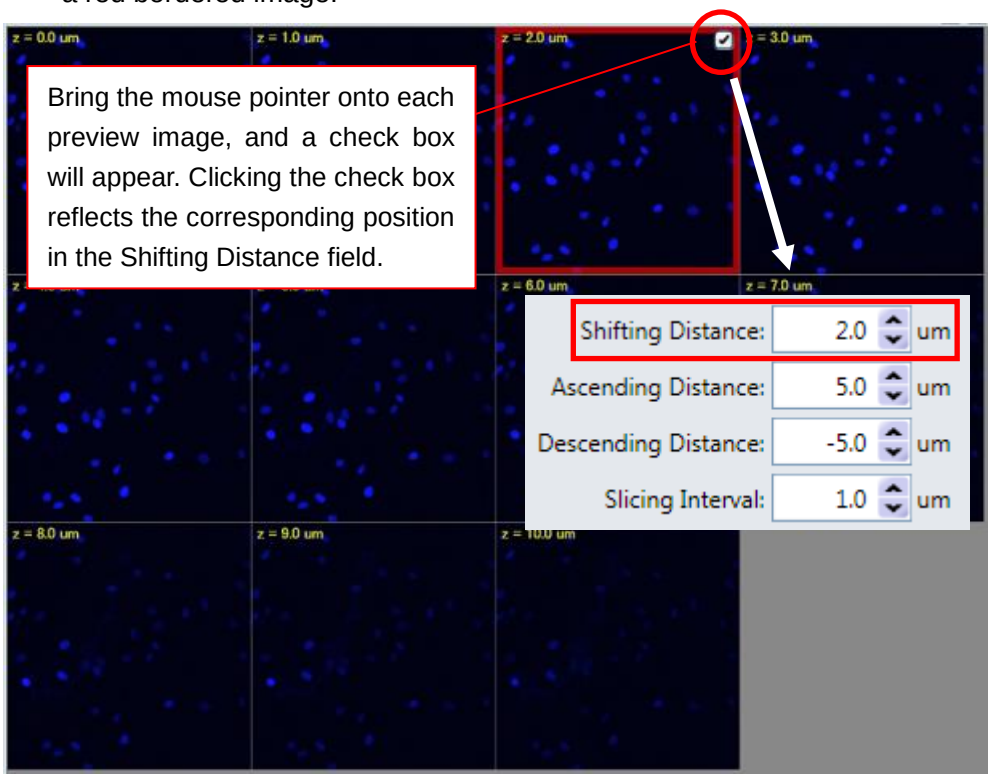
8) Click “Load Well Plate.” The well plate is transferred to the reader. (Refer to 8.1 for the setting of a plate.)



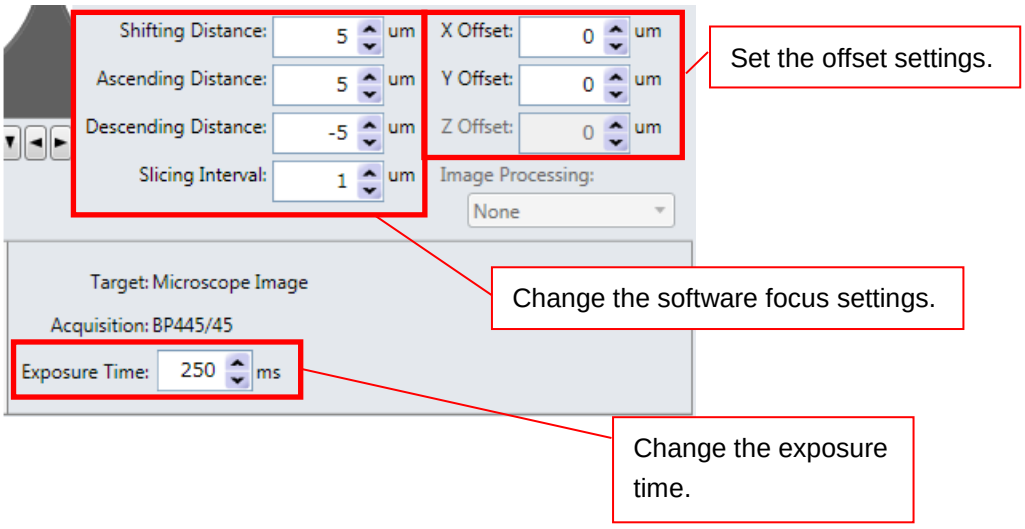
9) Click “Acquire Image” to display a preview.

10) A preview is displayed. (Refer to 5.9 and 6.2)

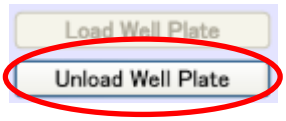
Bring the mouse pointer onto each preview image, and a check box will appear. Clicking the check box reflects the corresponding position in the Shifting Distance field, so click the check box of the image in best focus. The image of the highest brightness is shown with a red border, so look for a red bordered image.



11) After confirming the image selected on the preview screen, adjust the settings to appropriate values. If any of the settings has been changed, click “Acquire Image” to check the image on the preview screen again.



12) To remove the well plate, click “Unload Well Plate.” Remove the well plate from the stage. (Refer to 8.2)

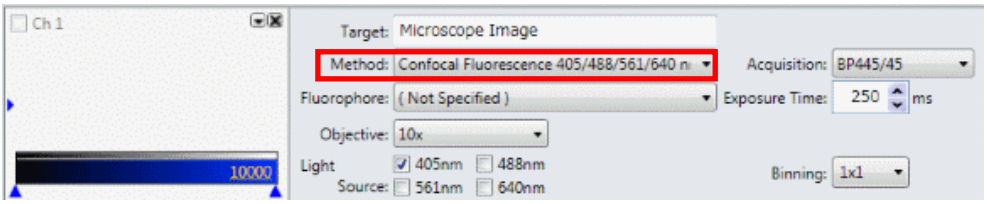


13) Click “OK.”

Setting Fluorescence Acquisition

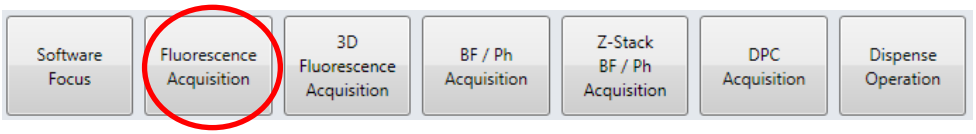
For the function of fluorescence acquisition, refer to 5.1.

1) Set the imaging channel. Under “Method,” select “Confocal Fluorescence” or “Epifluorescence.” (Refer to 5.5)

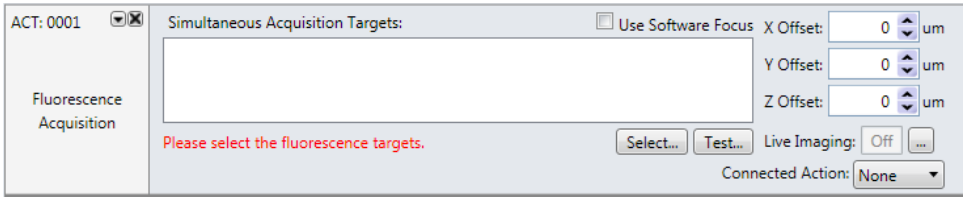


2) Click the Action List tab.

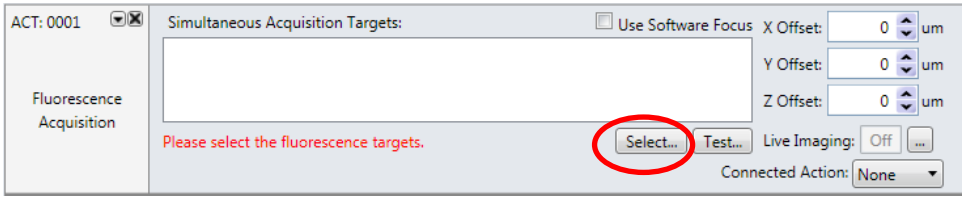
3) Click “Fluorescence Acquisition.”



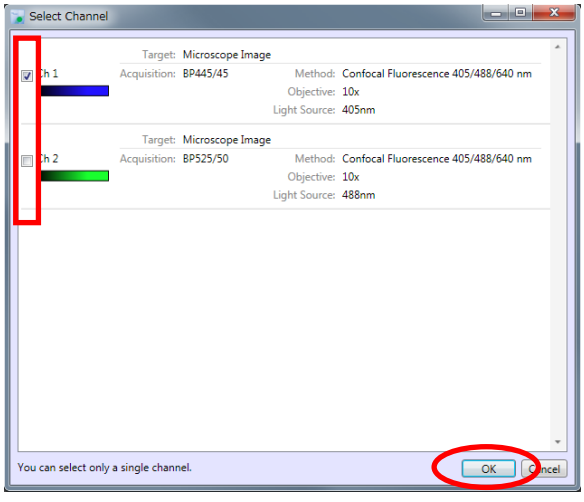
The screen for setting fluorescence imaging opens. (Refer to 6.2)



4) Click “Select."

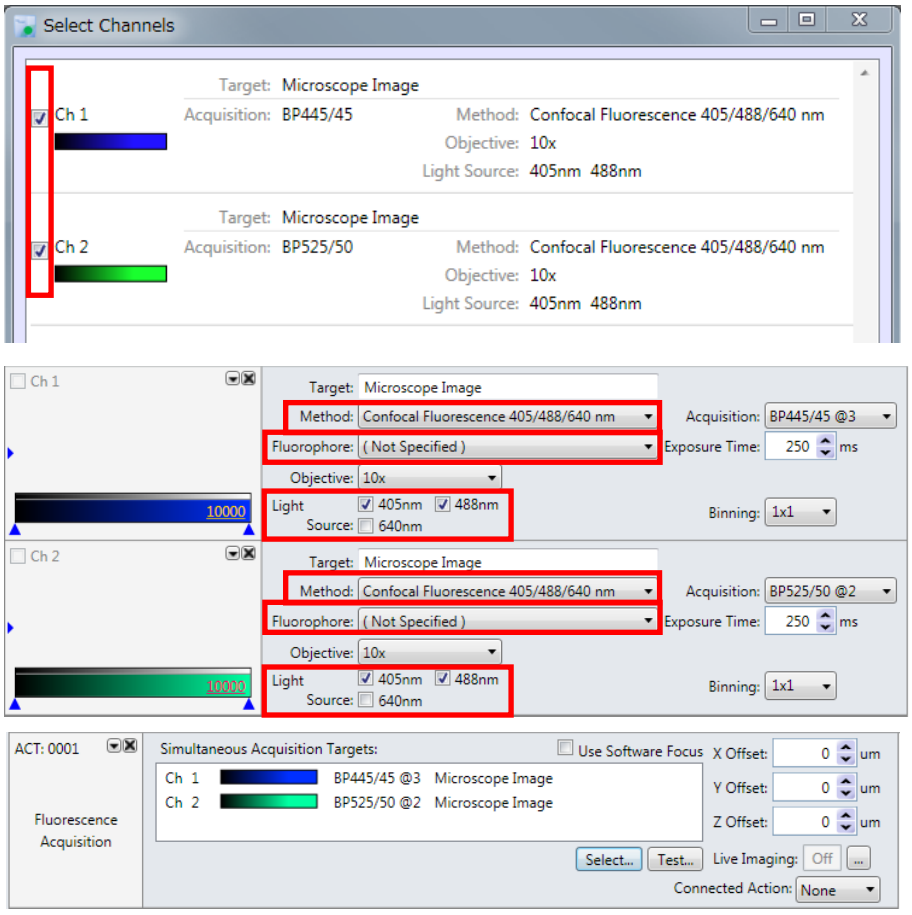


5) The Select Channels screen opens. Select the target channel, and then click “OK.” (Multiple channels can be selected.)

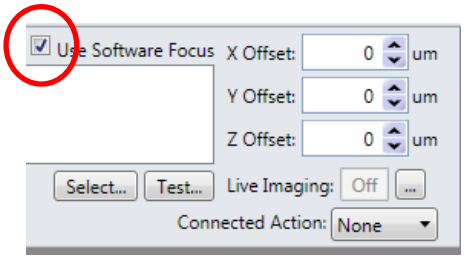


MEMO

- If multiple channels are selected on the Select Channels screen, laser beams of multiple wavelengths are emitted simultaneously.
- To emit laser beams of multiple wavelengths are emitted simultaneously, set the same optical system for all channels under “Method.”
- If emitting laser beams of multiple wavelengths simultaneously and performing crosstalk correction by Image Correction Software, select fluorophore. Refer to 5.15 about registration of fluorophore.

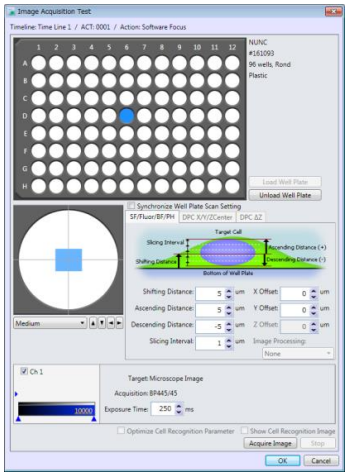


6) When “Use Software Focus” is checked, fluorescence images based on the software focused position are acquired. (Refer to 7.2)

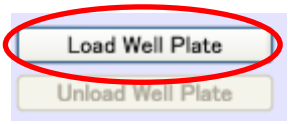


7) Click “Test.”

The Image Acquisition Test screen opens. (Refer to 6.2)

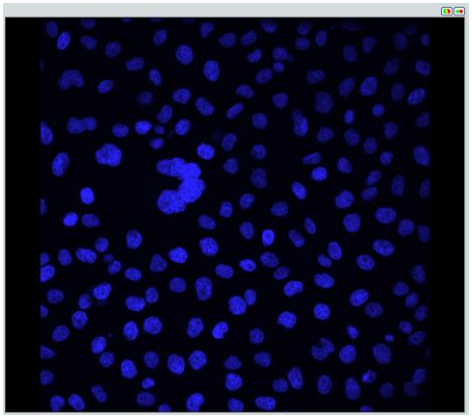


8) Click “Load Well Plate.” The well plate is transferred to the reader. (Refer to 8.1 for the setting of a plate.)

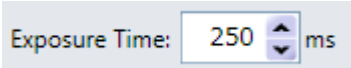


9) Click “Acquire Image” to display a preview.

10) The preview screen opens. (Refer to 5.9 and 6.2)
Change the value of Z Offset while checking the preview screen to set the best focal plane of cells.

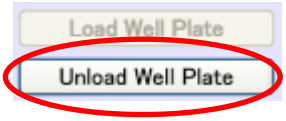


11) Adjust the exposure time.



12) After all items have been set, click “Stop” to stop the preview.

13) To remove the well plate, click “Unload Well Plate.” Remove the well plate from the stage. (Refer to 8.2)

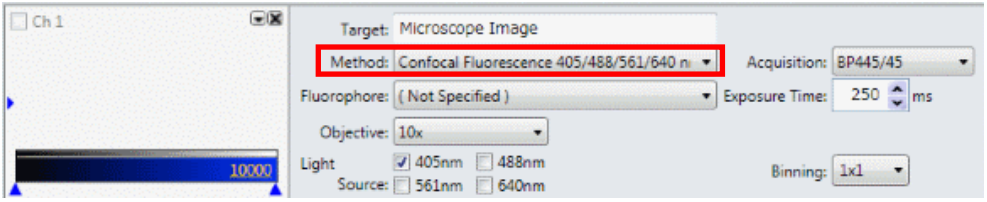


14) Click “OK.”

Setting 3D Fluorescence Acquisition

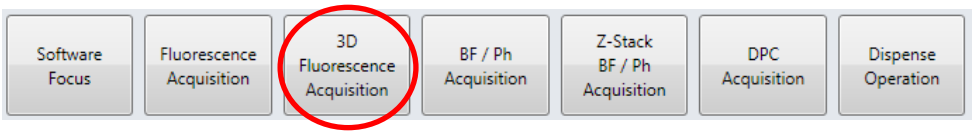
For the function of 3D fluorescence acquisition, refer to 5.1.

1) Set the imaging channel. Under “Method,” select “Confocal Fluorescence” or “Epifluorescence.”(Refer to 5.5)

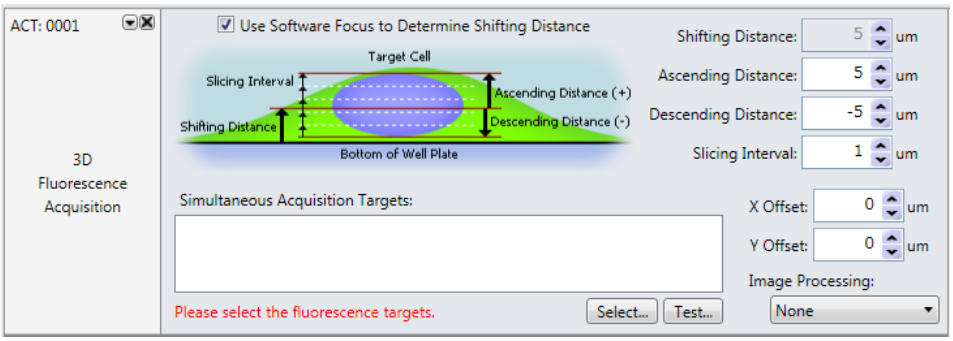


2) Click the Action List tab.

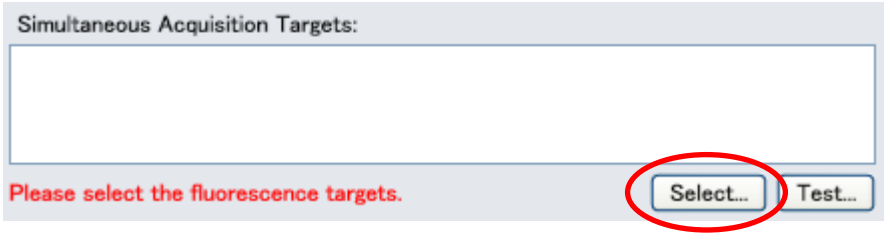
3) Click “3D Fluorescence Acquisition.”



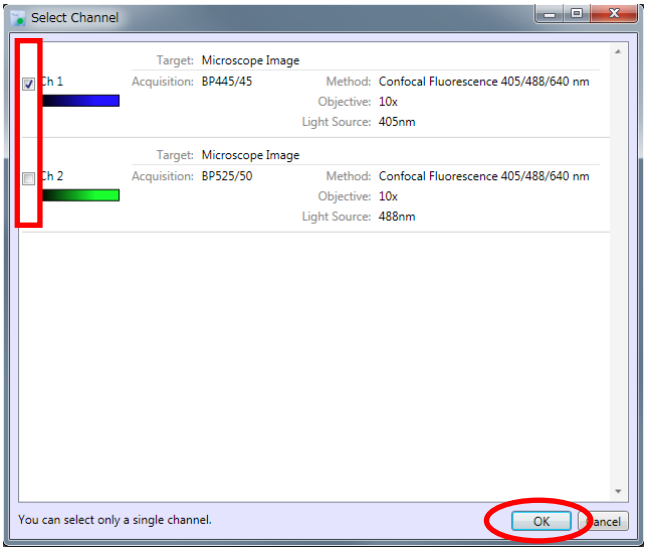
The screen for setting 3D fluorescence acquisition opens. (Refer to 6.2)



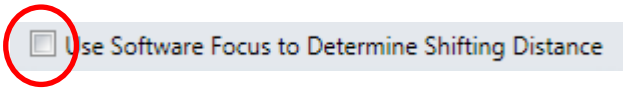
4) Specify the 3D imaging target.
Click “Select.”



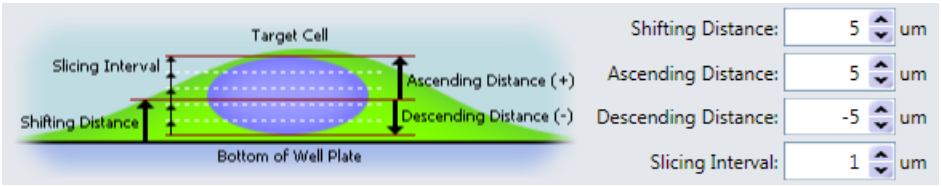
5) The Select Channel screen opens. Select the target channel, and then click "OK."



6) Uncheck "Use Software Focus to Determine Shifting Distance."



7) Set the 3D imaging area.

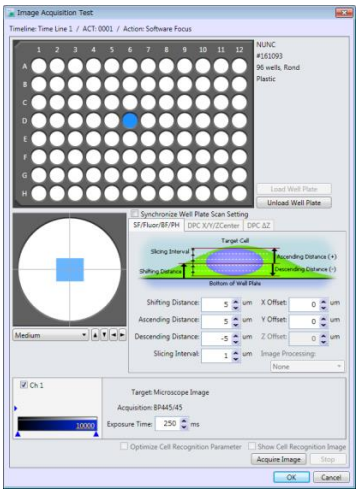


Item	Explanation
Shifting Distance	Amount of shift from the auto-focus position (Reference plane of 3D imaging)
Ascending Distance	Distance from the Shifting Distance position to the top plane of 3D imaging
Descending Distance	Distance from the Shifting Distance position to the bottom plane of 3D imaging
Slicing Interval	Z step width

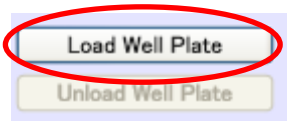
In the above example, the plane 5 µm above the auto-focus position is set as the reference plane of 3D imaging, and 11 images are captured in 1 µm steps over the area between 5 µm below and 5 µm above this focal plane.

8) Click "Test."

The Image Acquisition Test screen opens. (Refer to 6.2)



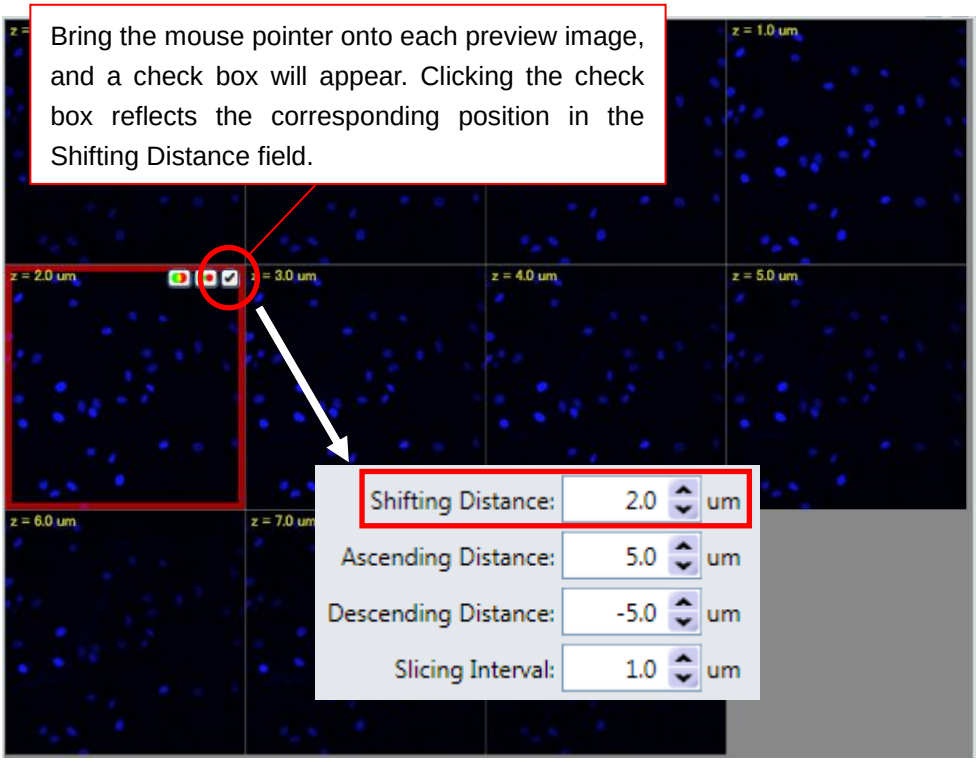
9) Click “Load Well Plate.” The well plate is transferred to the reader. (Refer to 8.1 for the setting of a plate.)



10) Click “Acquire Image” to display a preview.

11) A preview is displayed. (Refer to 5.9 and 6.2)

Bring the mouse pointer onto each preview image, and a check box will appear. Clicking the check box reflects the corresponding position in the Shifting Distance field, so click the check box of the image in best focus. The image of the highest brightness is shown with a red border, so look for a red bordered image.



12) An output method for captured Z images can be selected from the “Image Processing” items.

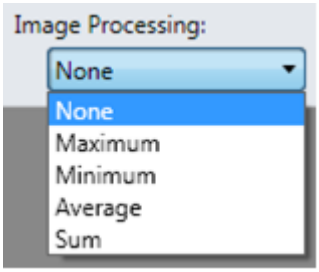
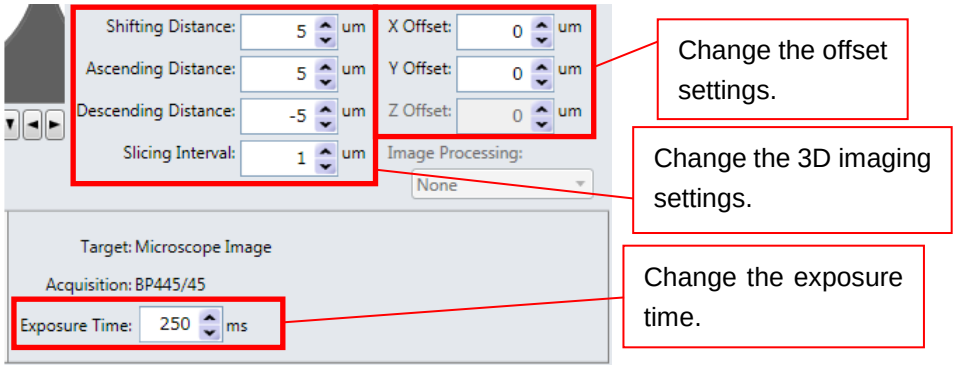


Image Processing	Explanation
None	Acquired each Z image is saved to output folder.
Maximum	MIP images are saved to output folder. (Refer to 5.1.)
Minimum	MinIP images are saved to output folder. (Refer to 5.1.)
Average	AIP images are saved to output folder. (Refer to 5.1.)
Sum	SUM images are saved to output folder. (Refer to 5.1)

13) Adjust the set values while checking the preview screen. If any of the settings has been changed, click “Acquire Image” to check the image on the preview screen again.

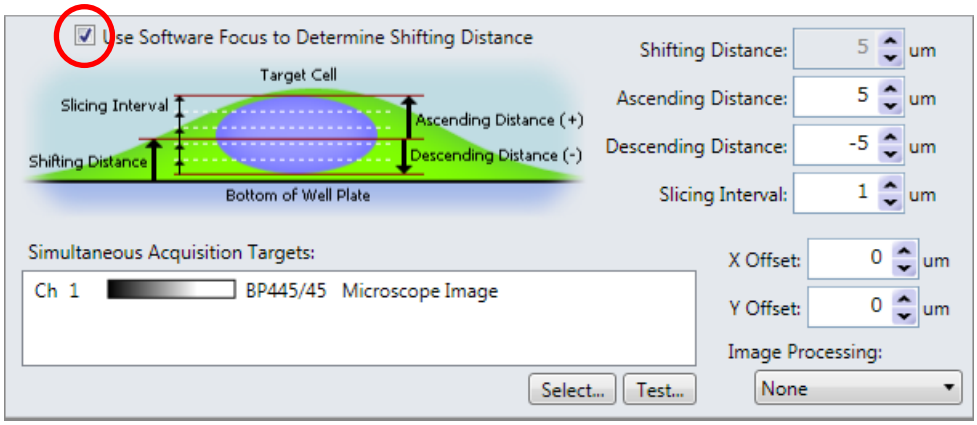


14) To remove the well plate, click “Unload Well Plate.” Remove the well plate from the stage. (Refer to 8.2)

15) Click “OK.”

MEMO

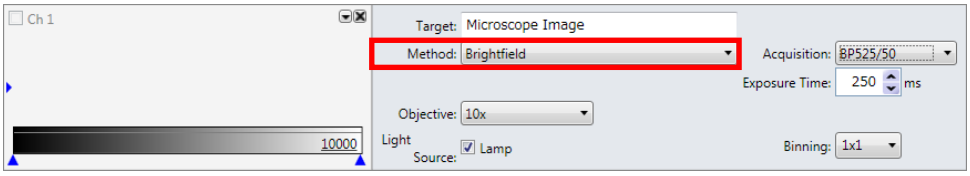
●Selecting the “Use Software Focus to Determine Shifting Distance” check box lets you set the Shifting Distance value for the software focus plane so that 3D imaging can be performed by using the software focus plane as a reference. (Refer to 7.2)



Setting Bright Field/Phase Contrast Acquisition

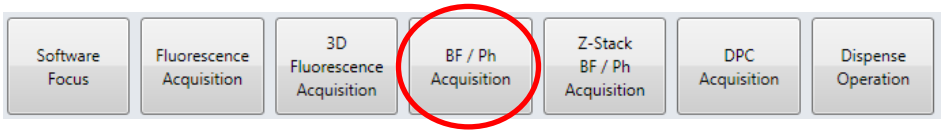
For the function of bright field/phase contrast acquisition, refer to 5.1.

1) Set the imaging channel. Under “Method,” select “Brightfield” or “Phase Contrast.” (Refer to 5.5)



2) Click the Action List tab.

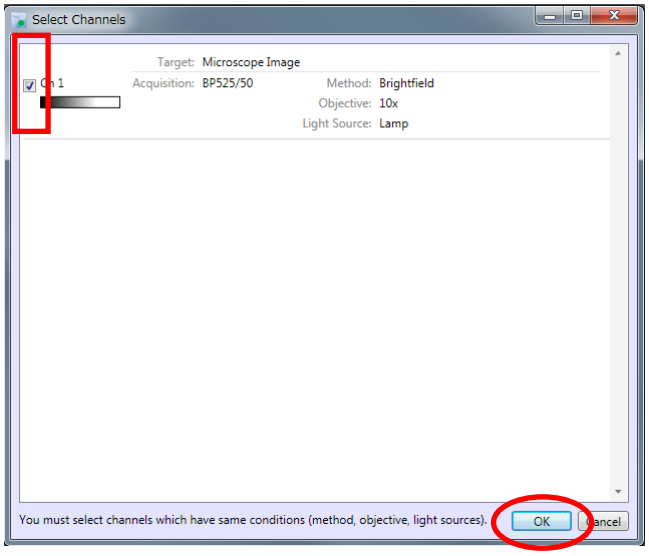
3) Click “Bright-field/Phase-contrast Acquisition.”



The screen for setting bright field/phase contrast imaging opens. (Refer to 6.2)

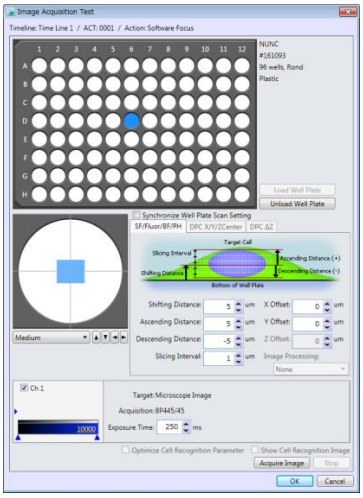
4) Click “Select.”

5) The Select Channel screen opens. Select the target channel, and then click “OK.”



6) Display an imaging preview to check the imaging settings. Click “Test.”

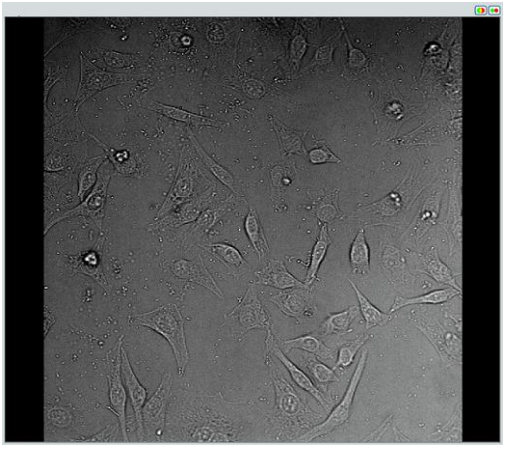
The Image Acquisition Test screen opens. (Refer to 6.2)

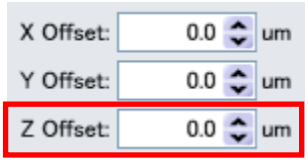


7) Click “Load Well Plate.” The well plate is transferred into the system. (Refer to 8.1 for the setting of a plate.)

8) Click “Acquire Image” to display a preview.

9) The preview screen opens. (Refer to 5.9 and 6.2)
Change the value of Z Offset while checking the preview screen to set the best focal plane of cells.

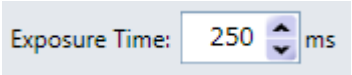




MEMO

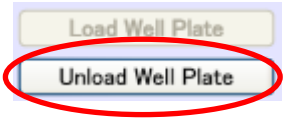
●In the phase contrast mode, correct phase-contrast images cannot be obtained except for the center of the well.

10) Adjust the exposure time.



11) After all items have been set, click “Stop” to stop the preview.

12) To remove the well plate, click “Unload Well Plate.” Remove the well plate from the stage. (Refer to 8.2)

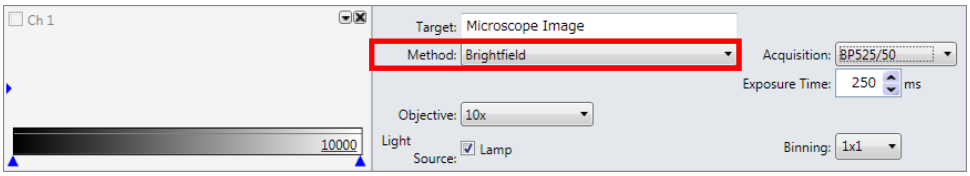


13) Click “OK.”

Setting Z-stack Bright-field/Phase-contrast Acquisition

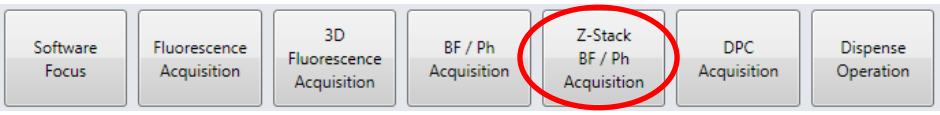
For the function of Z-stack bright-field/phase-contrast acquisition, refer to 5.1.

1) Set the imaging channel. Under “Method,” select “Brightfield” or “Phase Contrast.” (Refer to 5.5)

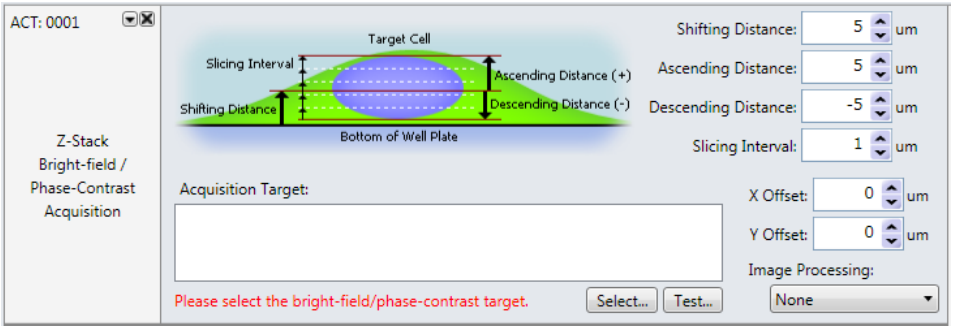


2) Click the Action List tab.

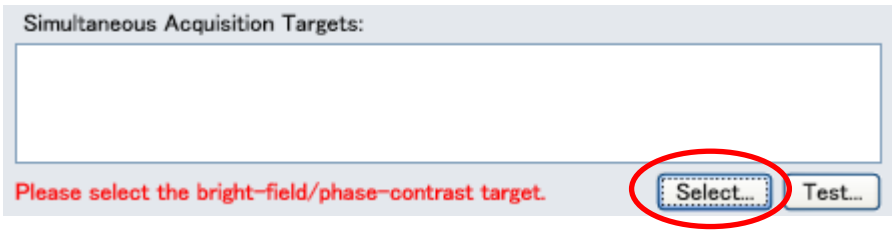
3) Click “Z-Stack Bright-field/Phase-contrast Acquisition.”



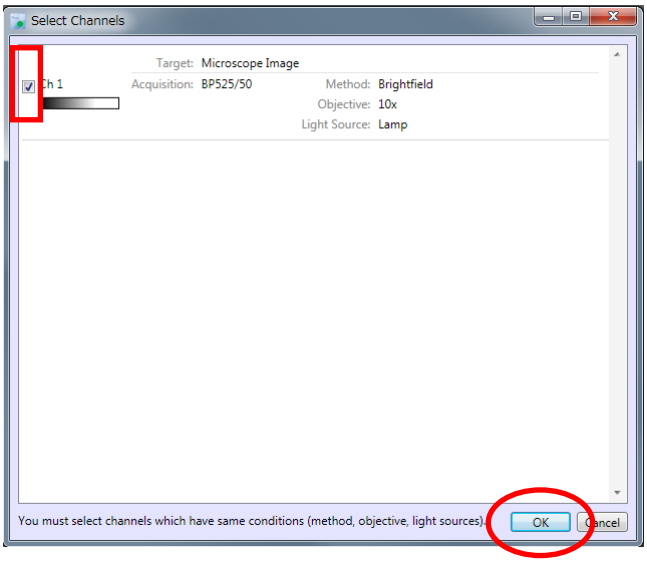
The screen for setting Z-stack bright-field/phase-contrast imaging opens. (Refer to 6.2)



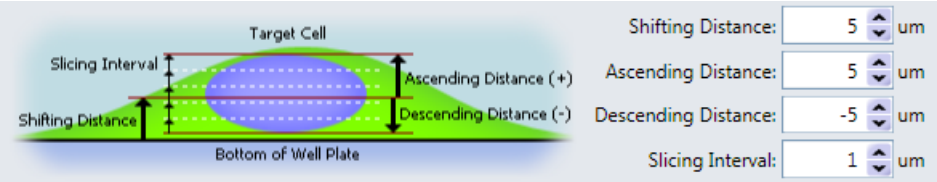
4) Click “Select.”



5) The Select Channel screen opens. Select the target channel, and then click “OK.”



6) Set the Z-stack imaging area.

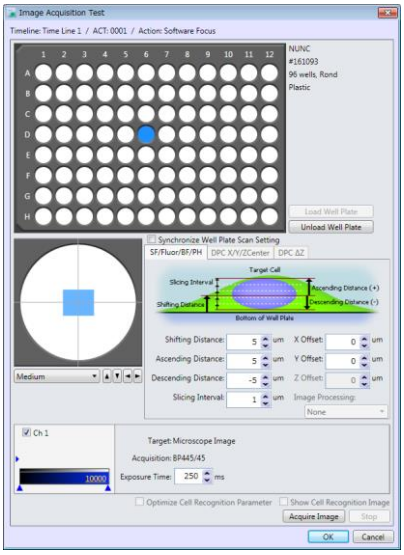


Item	Explanation
Shifting Distance	Amount of shift from the auto-focus position (Reference plane of Z-stack imaging)
Ascending Distance	Distance from the Shifting Distance position to the top plane of Z-stack imaging
Descending Distance	Distance from the Shifting Distance position to the bottom plane of Z-stack imaging
Slicing Interval	Z step width

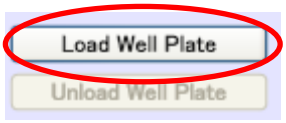
In the above example, the plane 5 µm above the auto-focus position is set as the reference plane of Z-stack imaging, and 11 images are captured in 1 µm steps over the area between 5 µm below and 5 µm above this focal plane.

7) Display an imaging preview to check the imaging settings. Click “Test.”

The Image Acquisition Test screen opens. (Refer to 6.2)

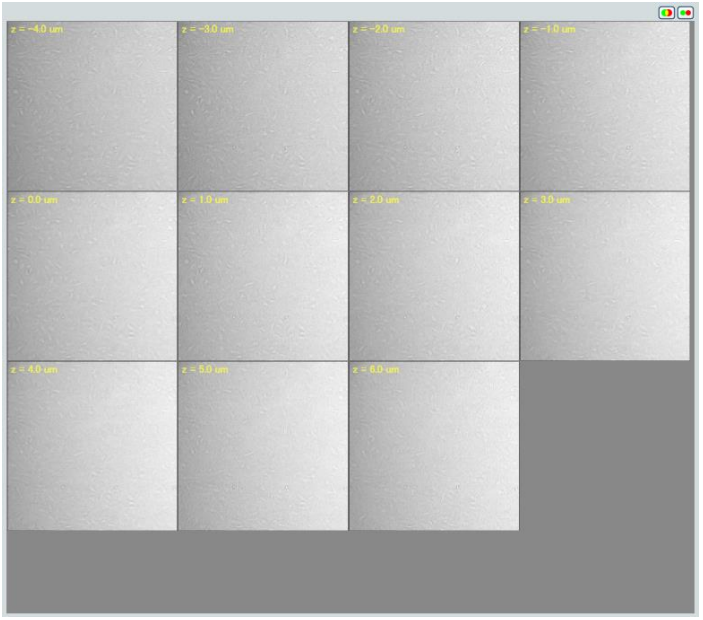


8) Click “Load Well Plate.” The well plate is transferred into the system. (Refer to 8.1 for the setting of a plate.)



9) Click “Acquire Image” to display a preview.

10) The preview screen opens. (Refer to 5.9 and 6.2.)



MEMO

●In the phase contrast mode, correct phase-contrast images cannot be obtained except for the center of the well.

11) An output method for captured Z images can be selected from the “Image Processing” items.

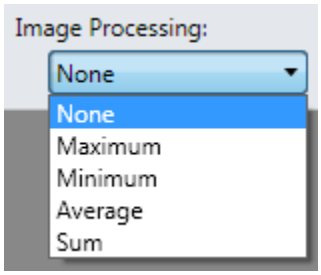
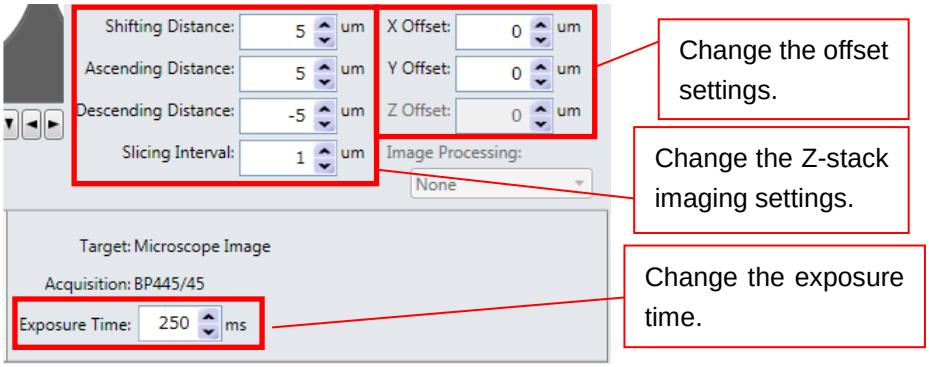


Image Processing	Explanation
None	Acquired each Z image is saved to output folder.
Maximum	MIP images are saved to output folder. (Refer to 5.1.)
Minimum	MinIP images are saved to output folder. (Refer to 5.1.)
Average	AIP images are saved to output folder. (Refer to 5.1.)
Sum	SUM images are saved to output folder. (Refer to 5.1)

12) Adjust the set values while checking the preview screen. If any of the settings has been changed, click “Acquire Image” to check the image on the preview screen again.



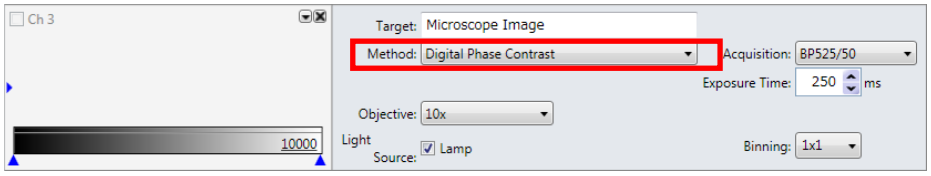
13) To remove the well plate, click “Unload Well Plate.” Remove the well plate from the stage. (Refer to 8.2)

14) Click “OK.”

Setting DPC Acquisition

Refer to 5.1 for the DPC acquisition function.

1) Set the imaging channel. Under “Method,” select “Digital Phase Contrast.” (Refer to 5.5)

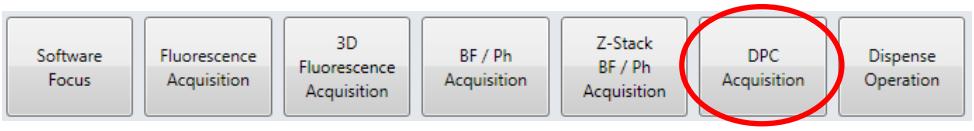


MEMO

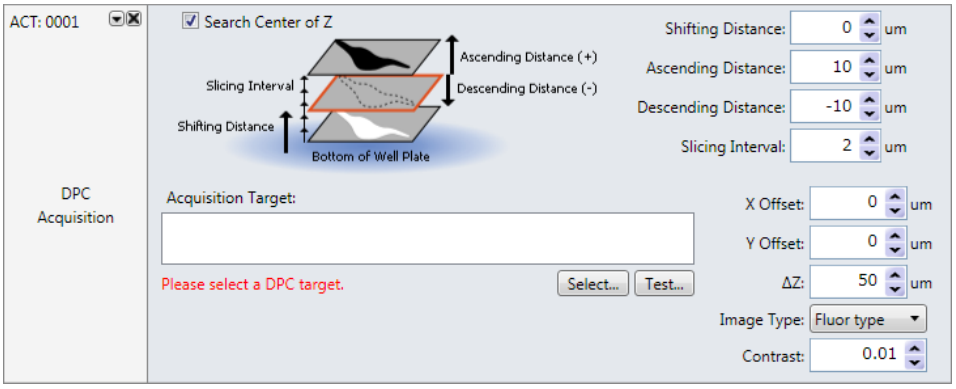
●Aberrant DPC image is generated from saturated image. Please adjust Lamp power and Exposure Time not to saturate.

2) Click the Action List tab.

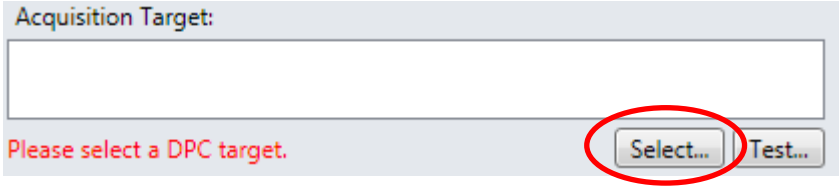
3) Click “DPC Acquisition”.



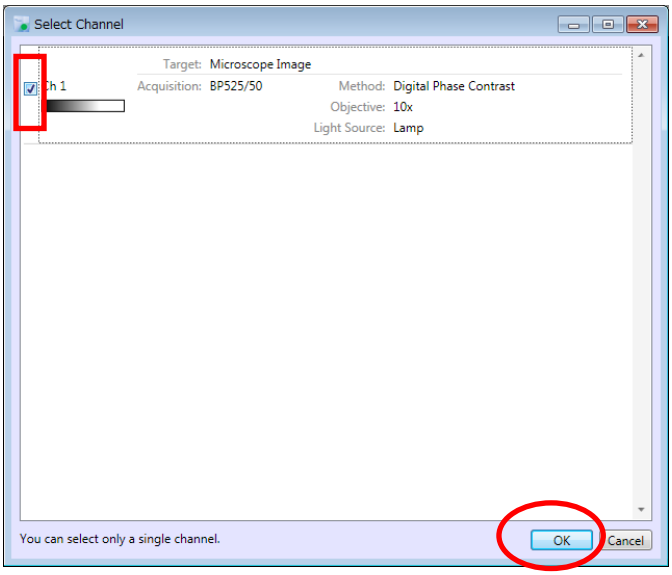
The screen for setting DPC Acquisition opens. (Refer to 6.2)



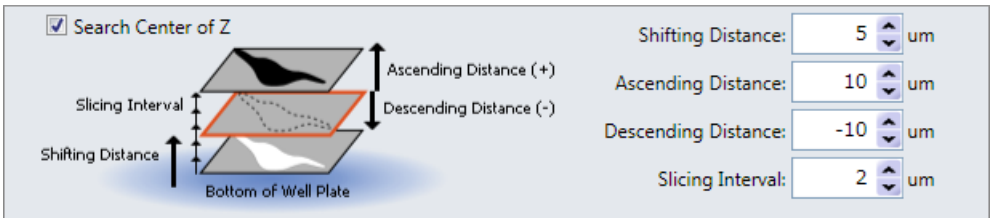
4) Click “Select”.



5) The Select Channel screen opens. Select the target channel, and then click "OK."



6) Set the automatic DPC reference position search.



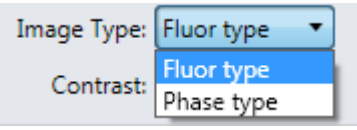
Item	Explanation
Search Center of Z	Check in case of performing automatic DPC reference position search.
Shifting Distance	Amount of shift from the auto-focus position (If "Search Center of Z" is checked, this Z position is reference plane of automatic DPC reference position search. If unchecked, this Z position is reference plane of DPC imaging)
Ascending Distance	Distance from the Shifting Distance position to the top plane of automatic DPC reference position search
Descending Distance	Distance from the Shifting Distance position to the bottom plane of automatic DPC reference position search
Slicing Interval	Z step width

MEMO

●DPC image is created from multiple images of different Z position by image processing. DPC reference position means the reference Z position of these multiple images.

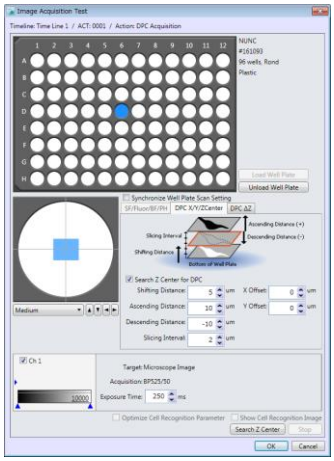
In the above example, the plane 5 µm above the auto-focus position is set as the reference plane of automatic DPC reference position search, and bright field images are captured in 2 µm steps over the area between 10 µm below and 10 µm above this focal plane. From the total of 11 images captured, the one with the most optimal for center of DPC acquisition is output as the image in focus.

7) Select DPC image type to output.

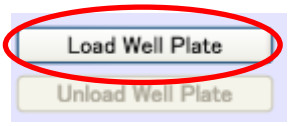


8) Display an imaging preview to check the imaging settings. Click “Test.”

The Image Acquisition Test screen opens. (Refer to 6.2)



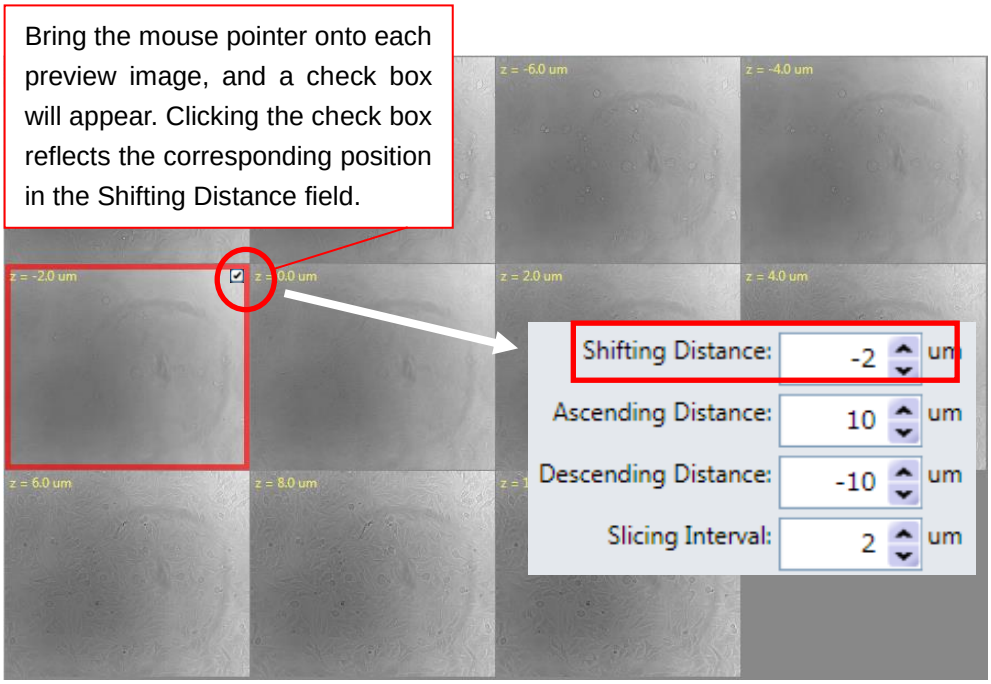
9) Click “Load Well Plate.” The well plate is transferred to the reader. (Refer to 8.1 for the setting of a plate.)



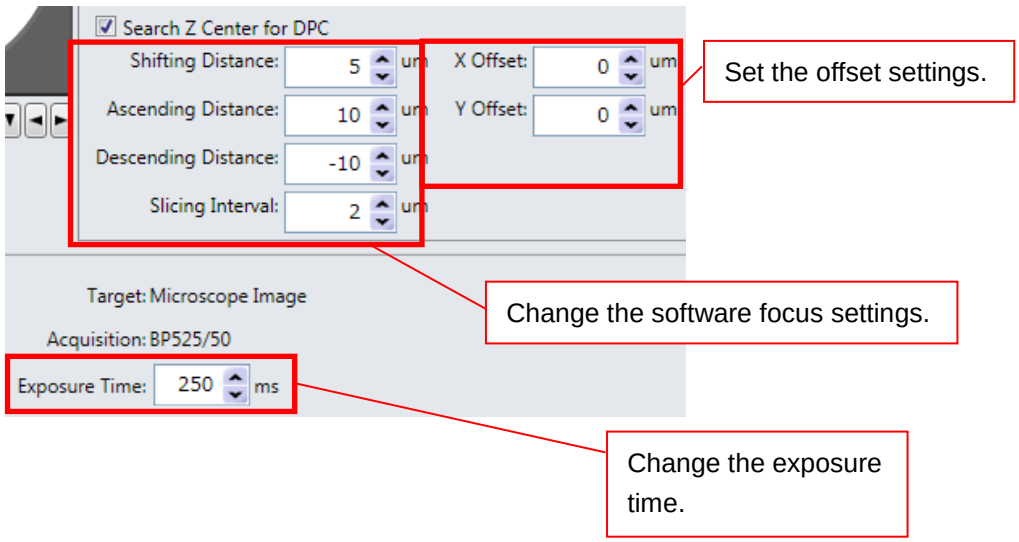
10) Click “Acquire Image” to display a preview.

11) A preview is displayed. (Refer to 5.9 and 6.2)

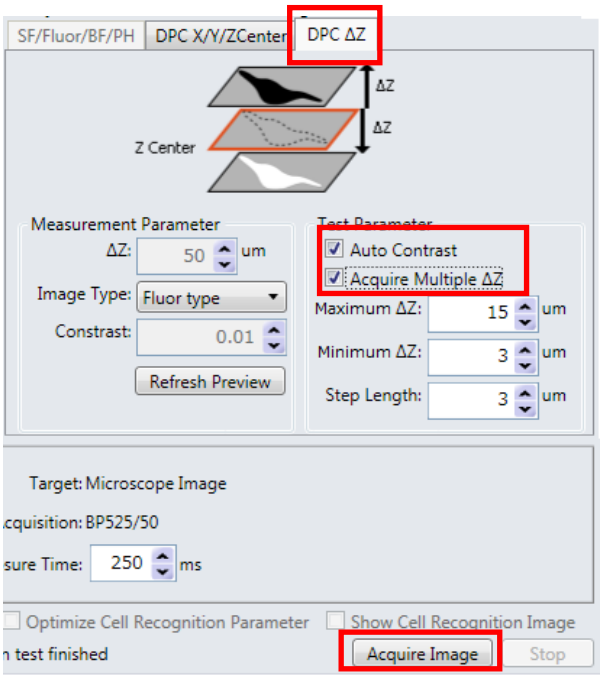
Bring the mouse pointer onto each preview image, and a check box will appear. Clicking the check box reflects the corresponding position in the Shifting Distance field, so click the check box of the image in best focus. The image of the most optimal for center of DPC acquisition is shown with a red border, so look for a red bordered image.



12) After confirming the image selected on the preview screen, adjust the settings to appropriate values. If any of the settings has been changed, click “Acquire Image” to check the image on the preview screen again.



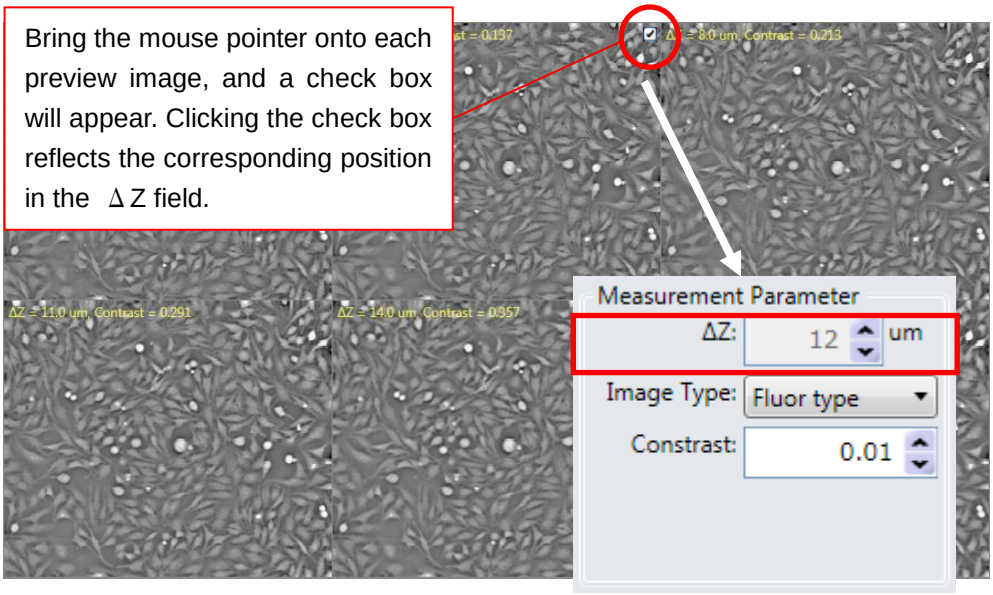
13) In case of changing DPC acquiring setting, select "ΔZ" tab. Check "Auto Contrast" and "Acquire Multiple ΔZ" and click "Acquire Image".



MEMO

●DPC image is created from multiple bright field images of different Z position by image processing. "ΔZ" in this procedure means distance of these Z positions.

14) A preview is displayed. (Refer to 5.9 and 6.2)
Bring the mouse pointer onto each preview image, and a check box will appear. Clicking the check box reflects the corresponding position in the ΔZ field, so click the check box of the image in the most optimal DPC image.



15) To remove the well plate, click “Unload Well Plate.” Remove the well plate from the stage. (Refer to 8.2)

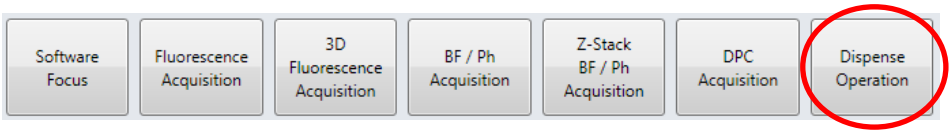
16) Click “OK.”

Setting Dispensing Operation

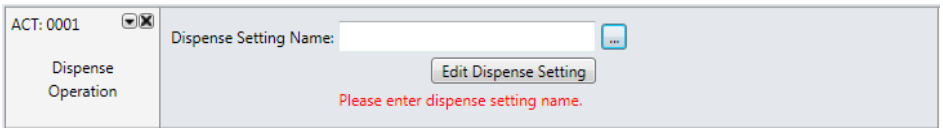
For the function of dispensing operation, refer to 5.1.

1) Click the Action List tab.

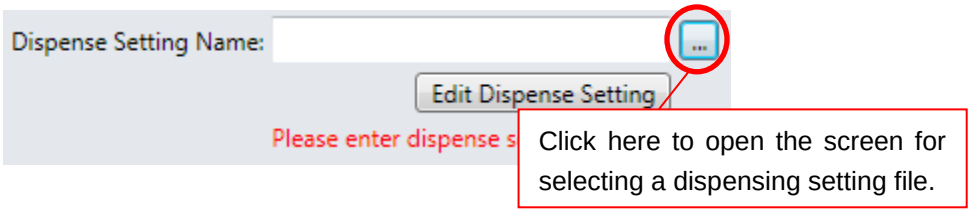
2) Click “Dispense Operation.”



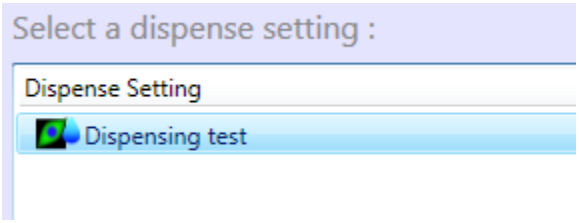
The screen for setting dispensing opens. (Refer to 6.2)

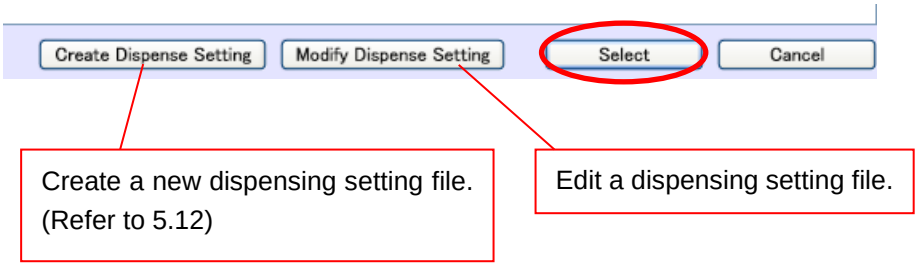


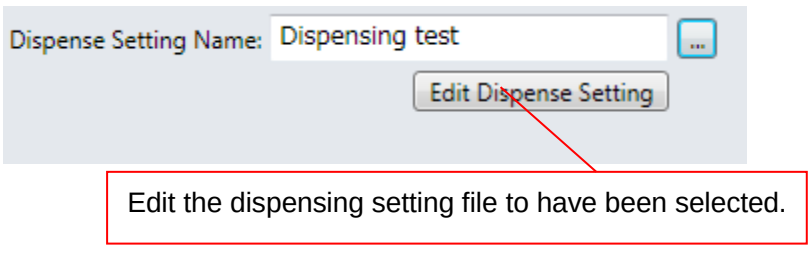
3) Select the dispensing setting file.



After selecting the dispensing setting file, click “Select.”





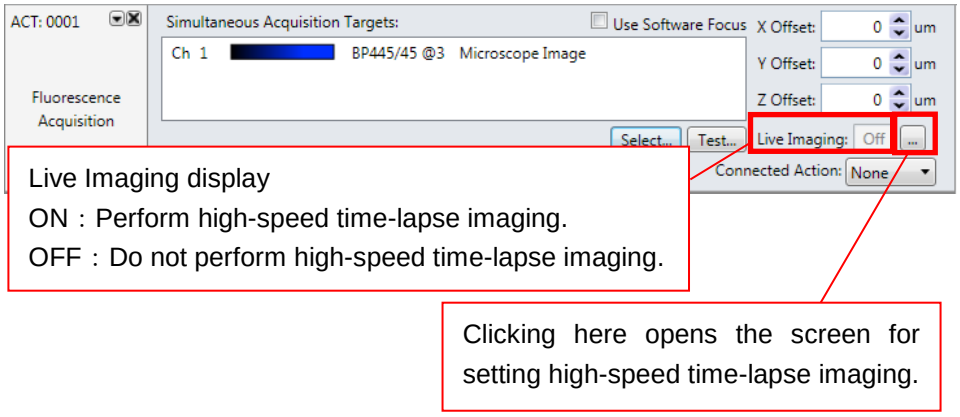


5.8. Setting High-speed Time-lapse Imaging

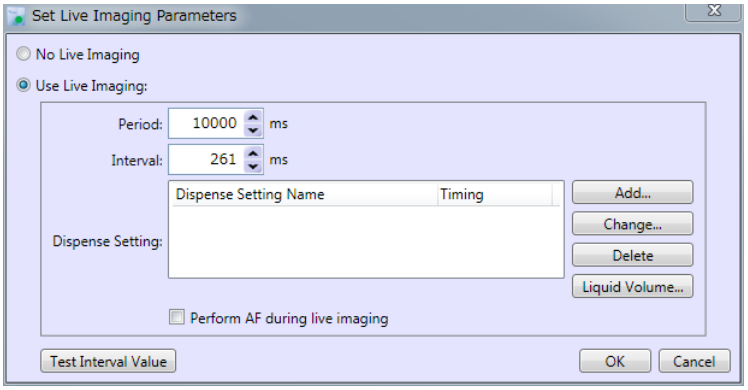
For the function of high-speed time-lapse imaging, refer to 5.1.

1) On the Action List tab, set a “Fluorescence Acquisition” task. (Refer to 5.7)

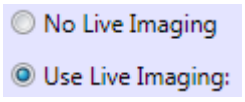
2) Open the screen for setting high-speed time-lapse imaging.



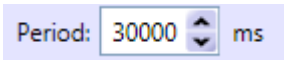
The screen for setting high-speed time-lapse imaging opens. (Refer to 6.2)



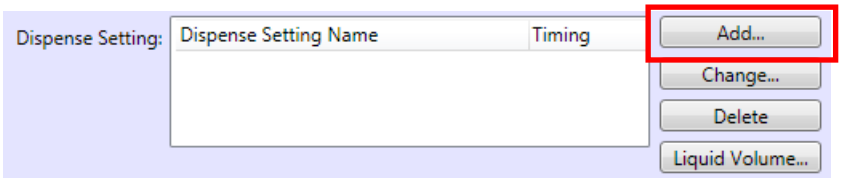
3) Select “Use Live Imaging.”

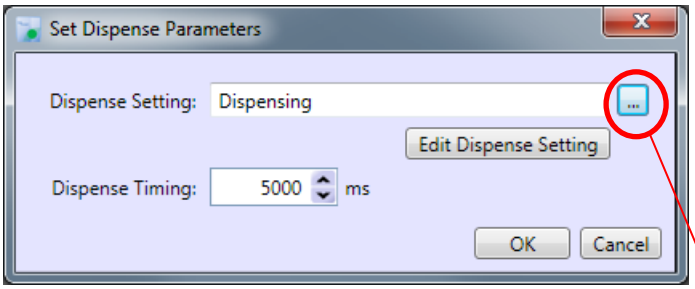


4) Enter the value of “Period” (period of high-speed time-lapse imaging).
(Interval≦Period)



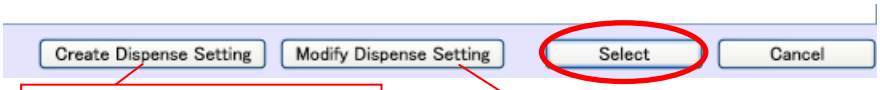
5) Set dispensing setting. (Set this item if dispensing is performed.)
Click “Add.”





Click to display the screen to select dispensing setting file.

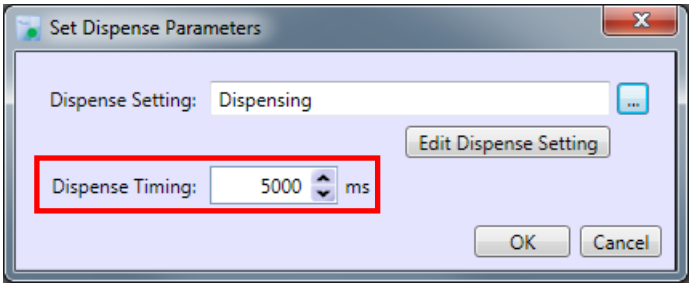
Select a dispensing setting file and then click “Select.”



Create a new dispensing setting file.
(Refer to 5.12 and 7.12.)

Edit a dispensing setting file.

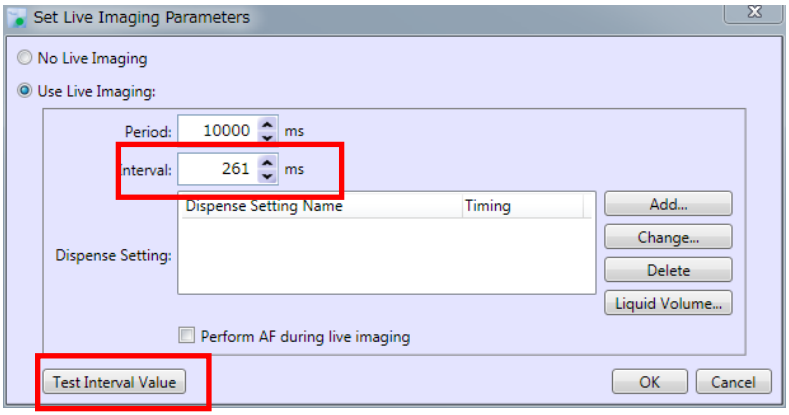
Enter the value of “Dispense Timing” (timing at which to drip reagent).
(1000ms≦Dispense Timing)



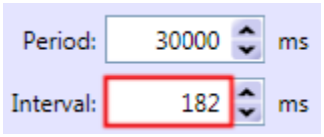
MEMO

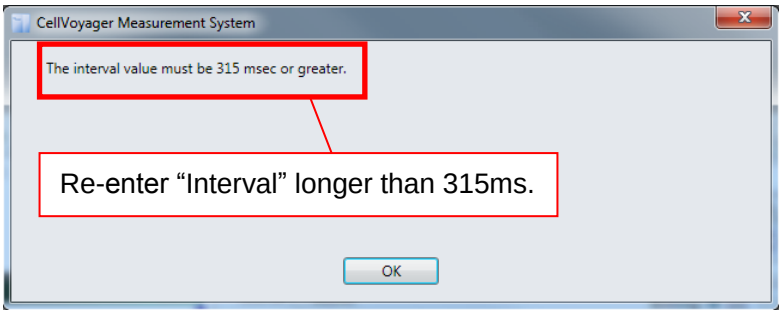
- Dispensing cannot be set for bright-field/phase-contrast acquisition.
- Up to three dispensing setting files can be assigned. (Refer to 5.1 for detail of multi dispensing.)

6) Enter the value of “Interval” (interval of high-speed time-lapse imaging). After entering, click “Test Interval Value.”



A message screen is displayed if the entered value for “Interval” is shorter than allowed. Re-enter “Interval” according to the message.

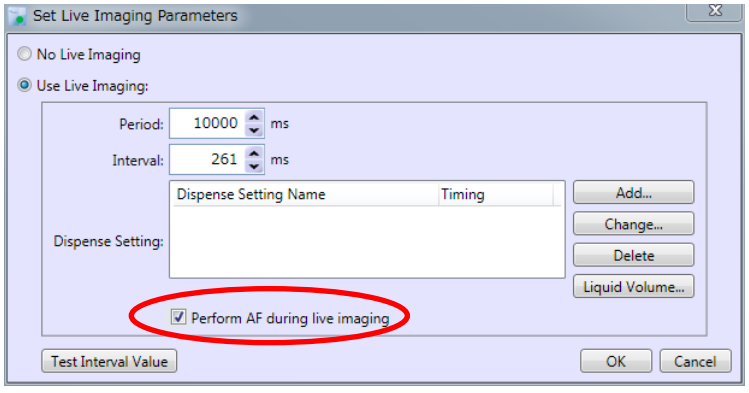




MEMO

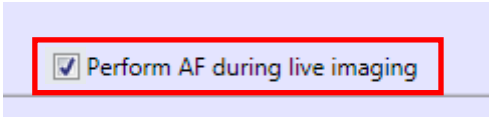
- Settable value for “Interval” is subject to some parameters such as exposure time, binning, transfer speed to storage, autofocus setting, etc.

7) Check “Perform AF during live imaging” to perform autofocus during high-speed time-lapse imaging.

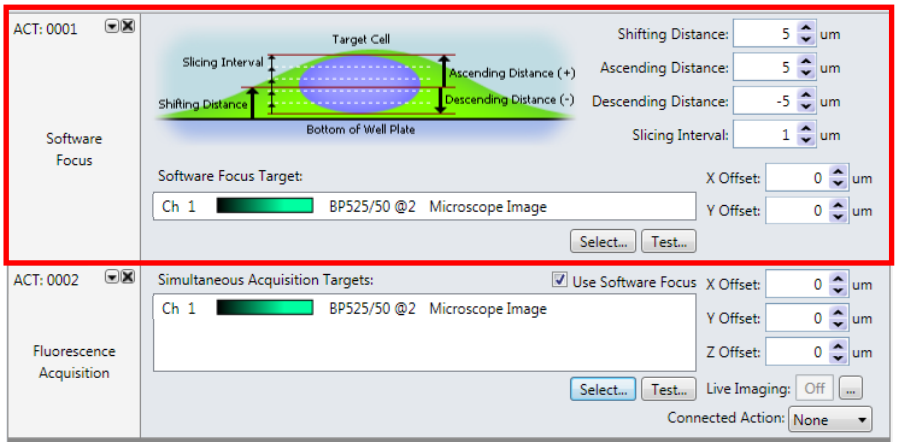


MEMO

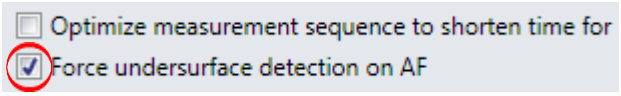
● Autofocus during high-speed time-lapse imaging cannot be performed if “Interval” isn’t long enough. (According to acquisition condition, about over than 5000 ms.)
In this case, red box appears around “Perform AF during live imaging”. Uncheck “Perform AF during live imaging” or set longer value to “Interval”.

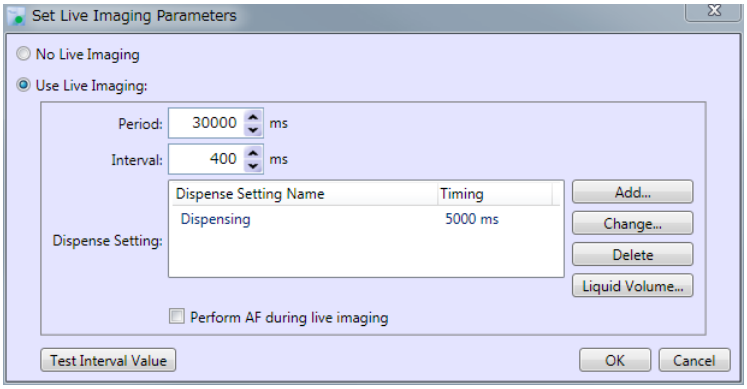


● Following setting is recommended when autofocus during high-speed time-lapse imaging is performed. Also, following setting must be set in combination.
- Set software focus at previous “Action List”.



- Check “Force undersurface detection on AF” at bottom of “Action List”.

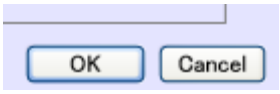


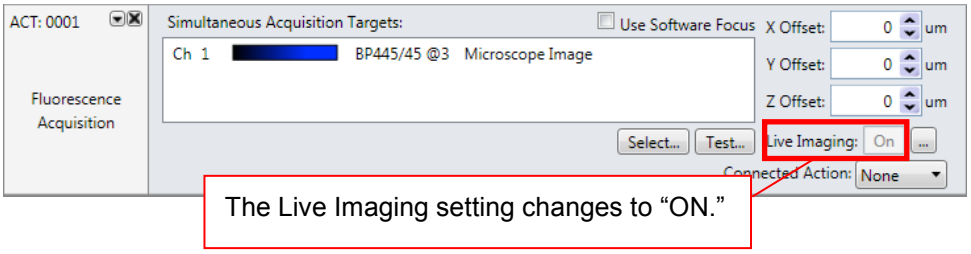


Example of a high-time lapse setting

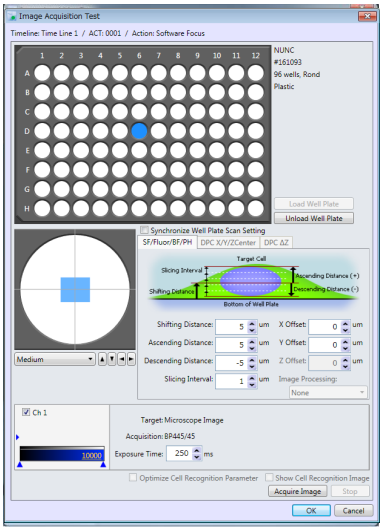
In the above example, time-lapse imaging is performed at an imaging interval of 400 ms and reagent is dropped 5 seconds after the start of imaging. The system moves to the next well after capturing for a period of 30 seconds.

8) After all items have been set, click “OK.”



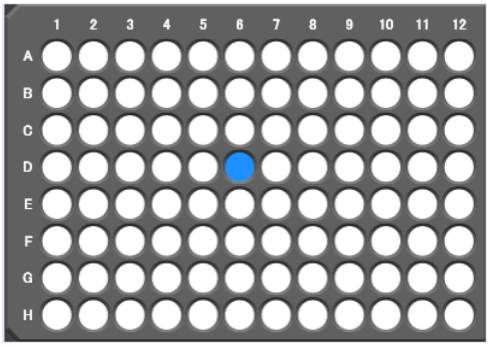


5.9. Setting the Preview Screen

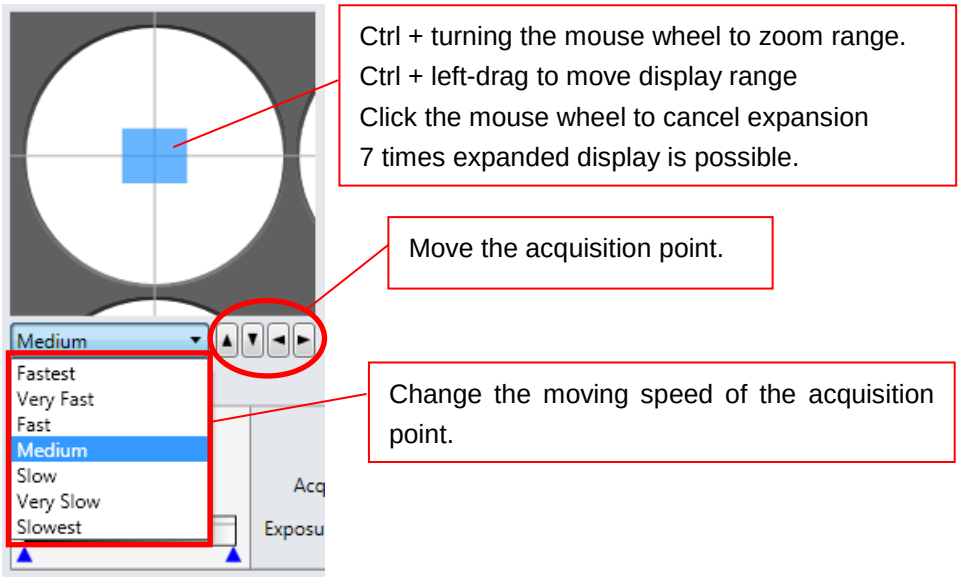


Preview screen (Refer to 6.2)

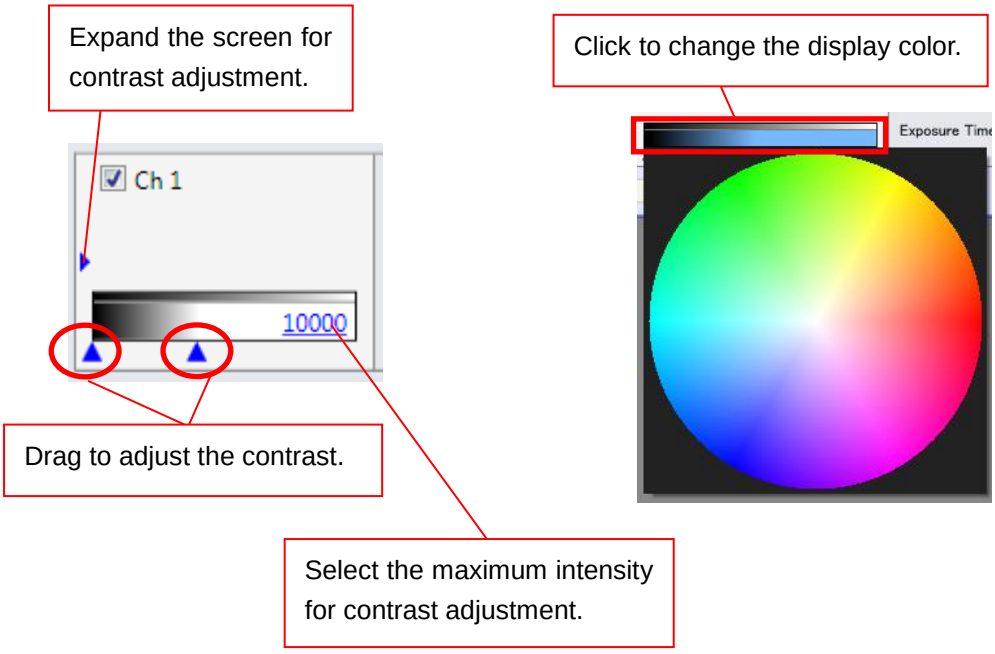
1) Select the well whose preview will be displayed.
   Click a desired well to be used for the image-acquisition test.



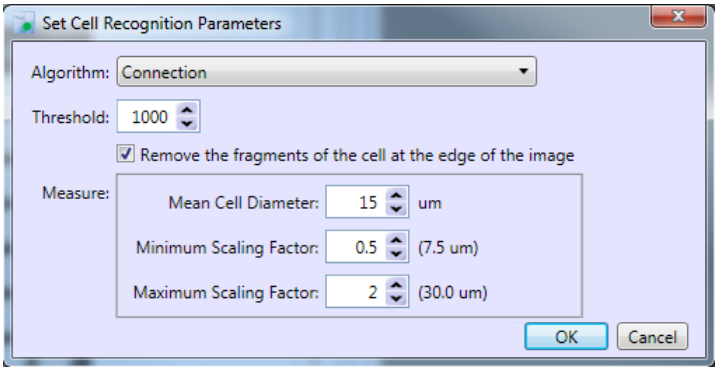
2) Specify the acquisition point in the well. Click the arrow buttons to move the acquisition point. Click on the well, and the clicked point will be specified as the imaging point.



3) Adjust the contrast on the preview screen. Click the contrast bar to change the display color for preview.



5.10. Setting the Cell Recognition Algorithm



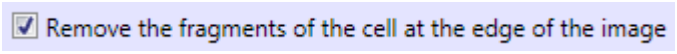
Cell recognition algorithm setting screen

Items displayed on the cell recognition algorithm setting screen

Item		Explanation
Algorithm		Sets the algorithm to count the number of cells.
	Binary per Mean Area	Algorithm to obtain the number of cells by binarizing cell images and dividing the total area of the recognized cells by the mean cell size.
	Connection	Algorithm to binarize cell images, perform cell labeling, and count the number of cells that fall within the range between the entered minimum and maximum scaling factors relative to the cell mean size.
	Connection and Binary per Mean Area	Algorithm to add the number of cells obtained by the Connection algorithm and the number of cells obtained by the Binary per Mean Area algorithm in relation to the number of cells that exceeds the maximum scaling factor relative to the cell mean size.
Threshold		Threshold to binarize cell images.
Remove the fragments of the cell at the edge of the image		Check the checkbox to exclude the cells at the edges of the image in the cell count.
Mean Cell Diameter		Specifies the mean cell diameter. Unit : µm
Minimum Scaling Factor		Sets the minimum scaling factor.
Maximum Scaling Factor		Sets the maximum scaling factor.

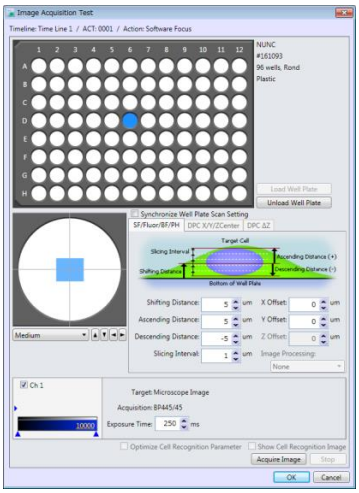
1) Select the cell recognition algorithm.

2) Select the following check box if you do not want the cells at edges of the image to be recognized.

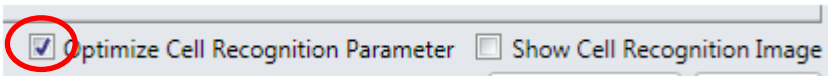


3) Do not set anything under “Threshold” and “Measure,” and click “OK.”

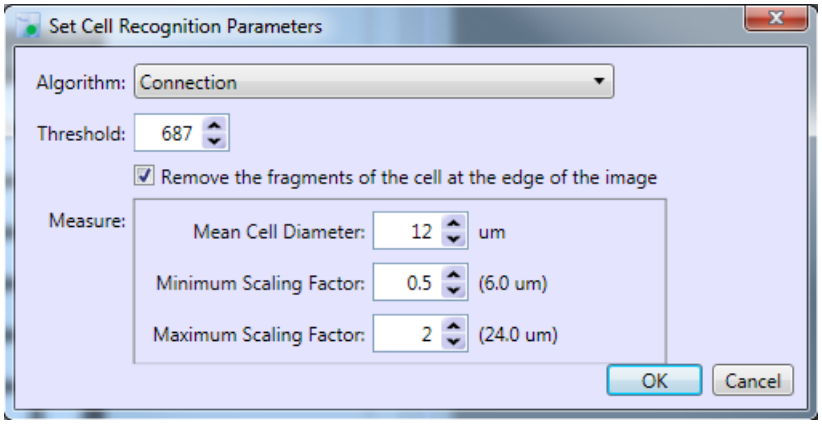
4) The preview screen opens based on the Action List tab settings to which the cell recognition algorithm has been applied. Adjust the well position, imaging position, etc. (Refer to 5.9)



5) Select the “Optimize Cell Recognition Parameter” (automatically optimize the set values of the recognition algorithm) checkbox, and then click “Acquire Image.”

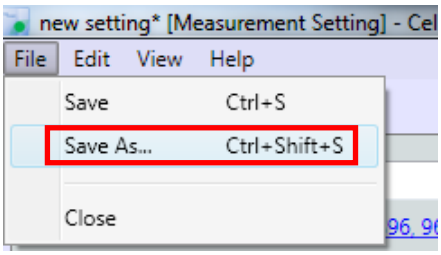


6) After checking the preview screen, click “Stop.” When the preview screen closes, the “Threshold” and “Measure” parameters on the cell recognition algorithm setting screen have been automatically set to optimal values. To manually set the parameters, unselect the “Optimize Cell Recognition Parameter” check box and adjust the “Threshold” and “Measure” parameters while checking the preview screen.

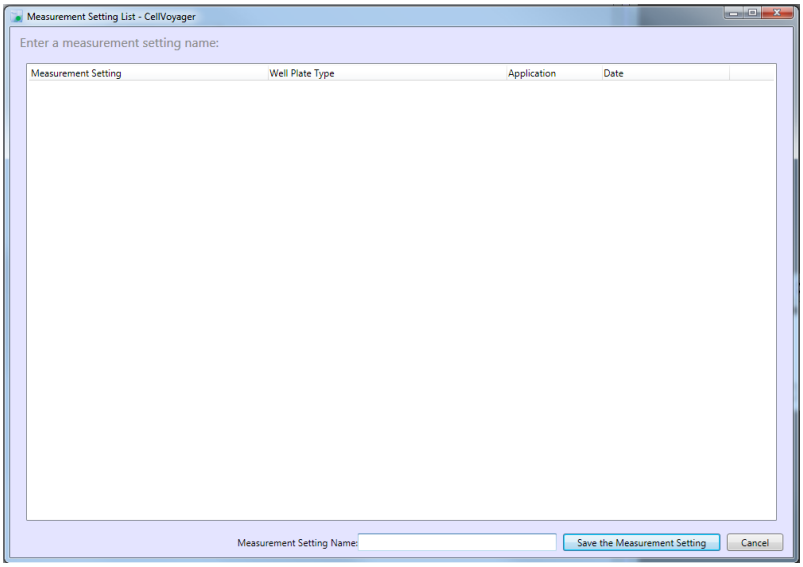


5.11. Saving the Measurement Setting File

1) In the menu of the measurement setting file, click “File” and then select “Save As.”

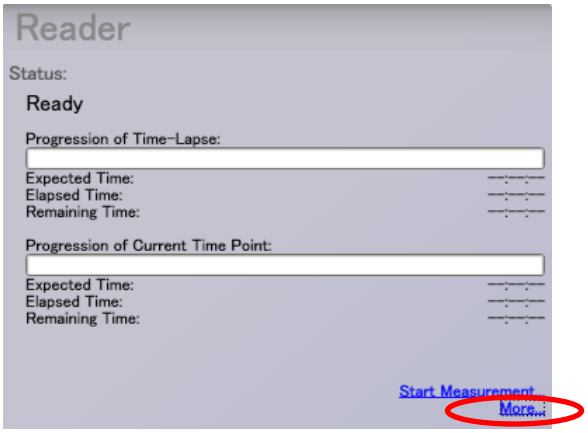


2) Enter the name under which to save the measurement setting file, and click “Save the Measurement Setting.”

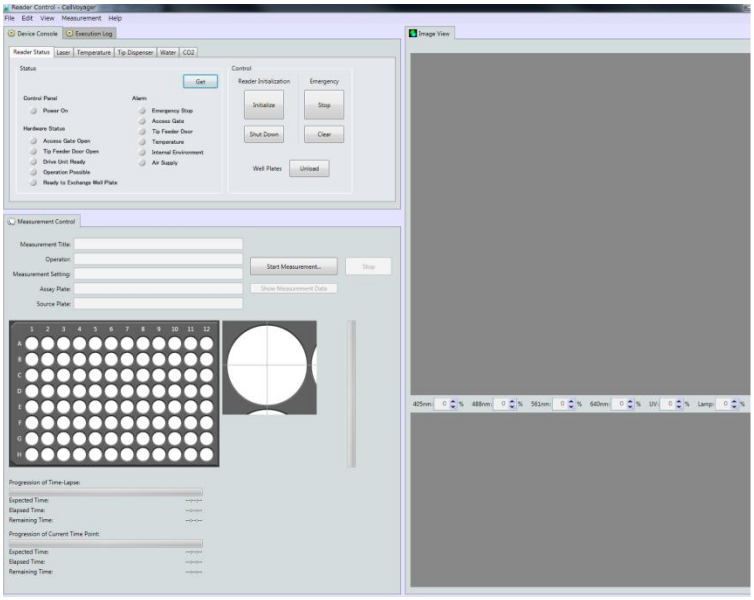


5.12. Creating a Dispensing Setting File

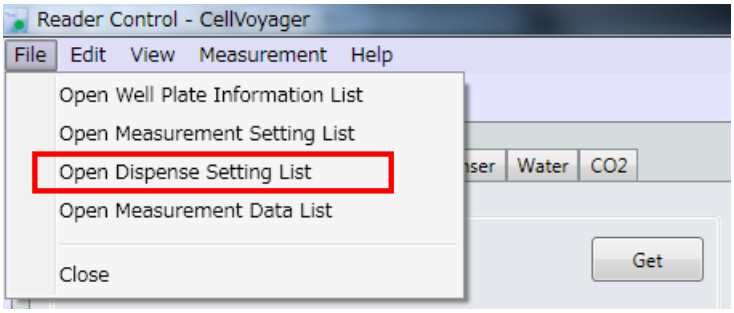
1) Click “More” in the Reader area to open the Reader Control screen.



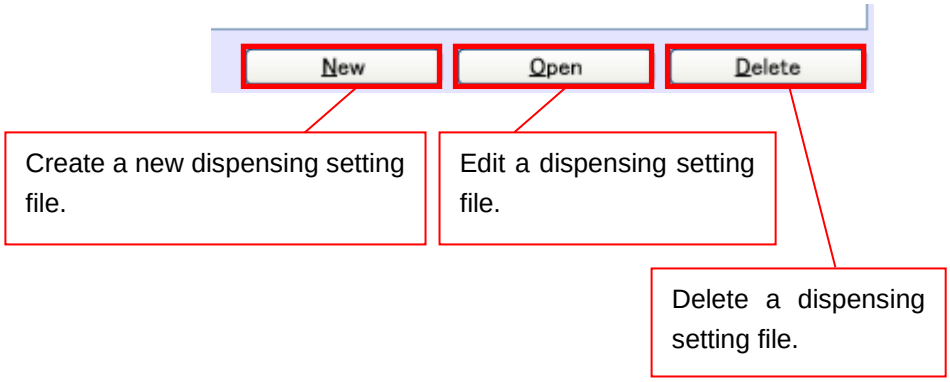
The Reader Control screen opens. (Refer to 6.4)



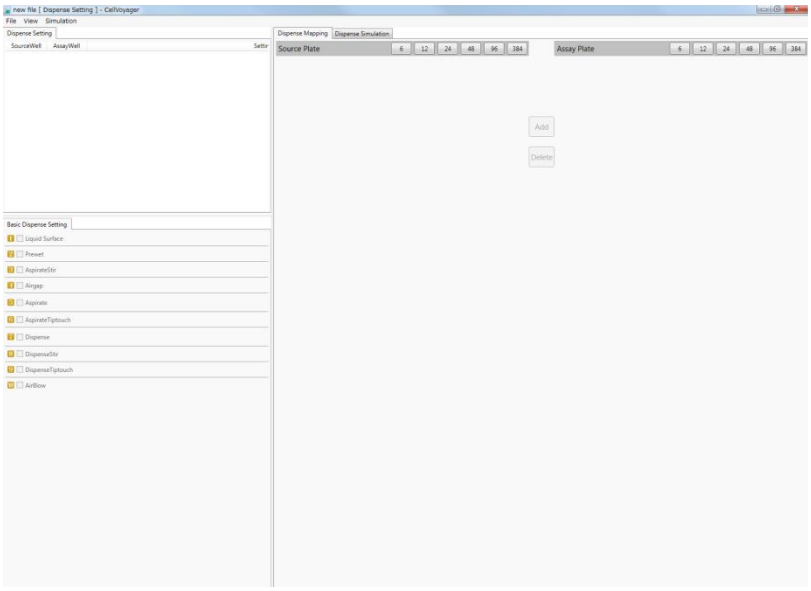
2) Click “Open Dispense Setting List” in the File menu.



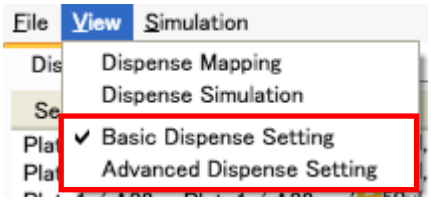
3) The Dispense Setting List screen opens. Click “New.”



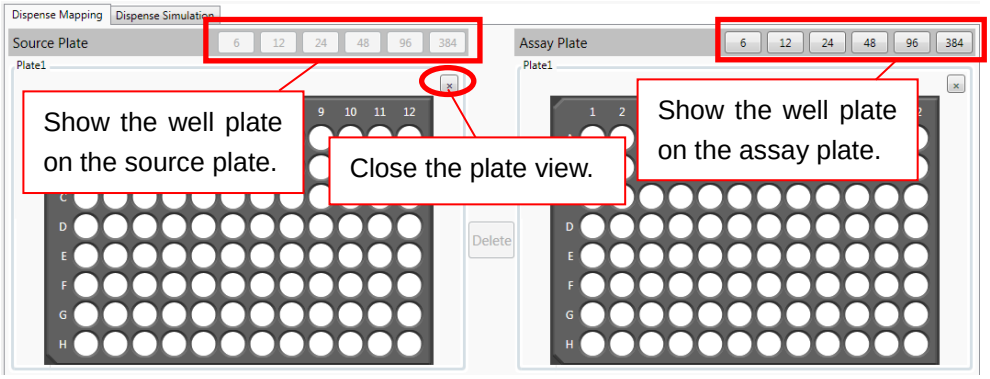
The Dispense Setting screen appears. (Refer to 6.3)



4) In the menu, click View -> Basic Dispense Setting or Advanced Dispense Setting, and select the dispensing setting mode. To configure the advanced setting for the dispensing operation, select Advanced Dispense Setting.



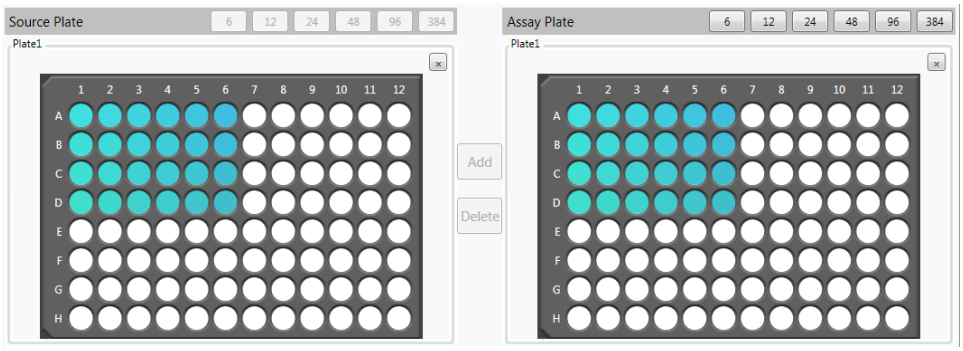
5) Display the Dispense Mapping tab and then display source plate and assay plate.



MEMO

- Only one source plate and up to four assay plates can be set.
- Dispensing to multiple assay plates is possible when a large incubator (sold separately) is used.

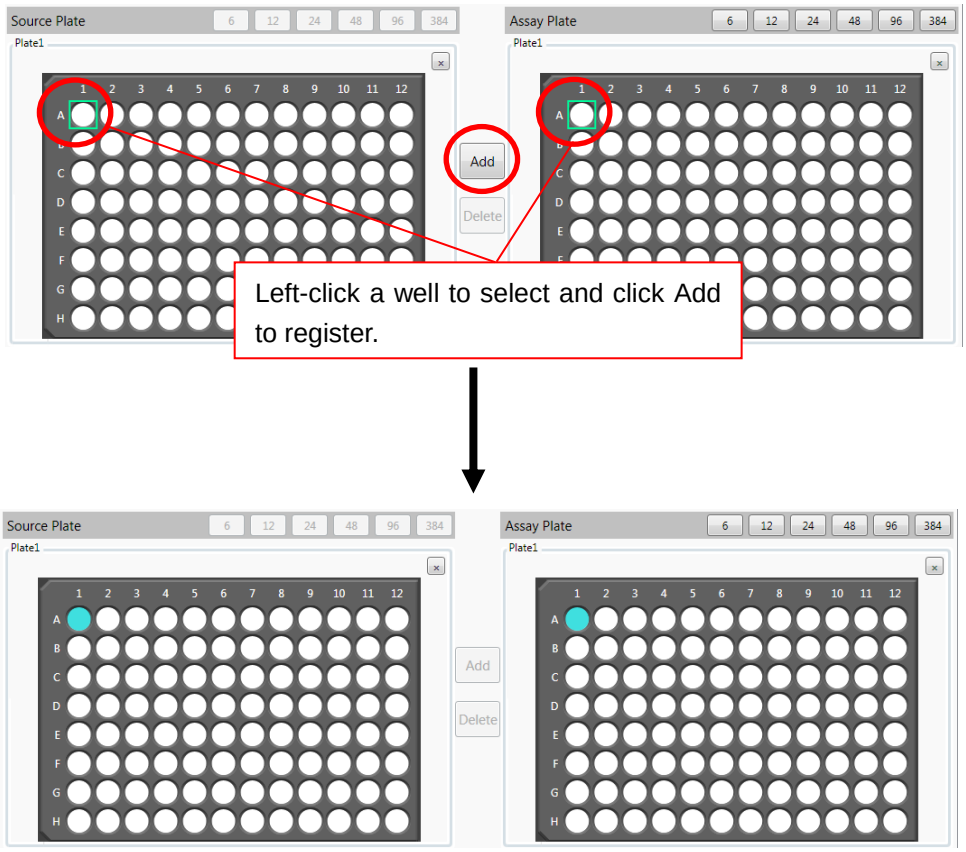
6) Link the wells on the source plate and assay plate.



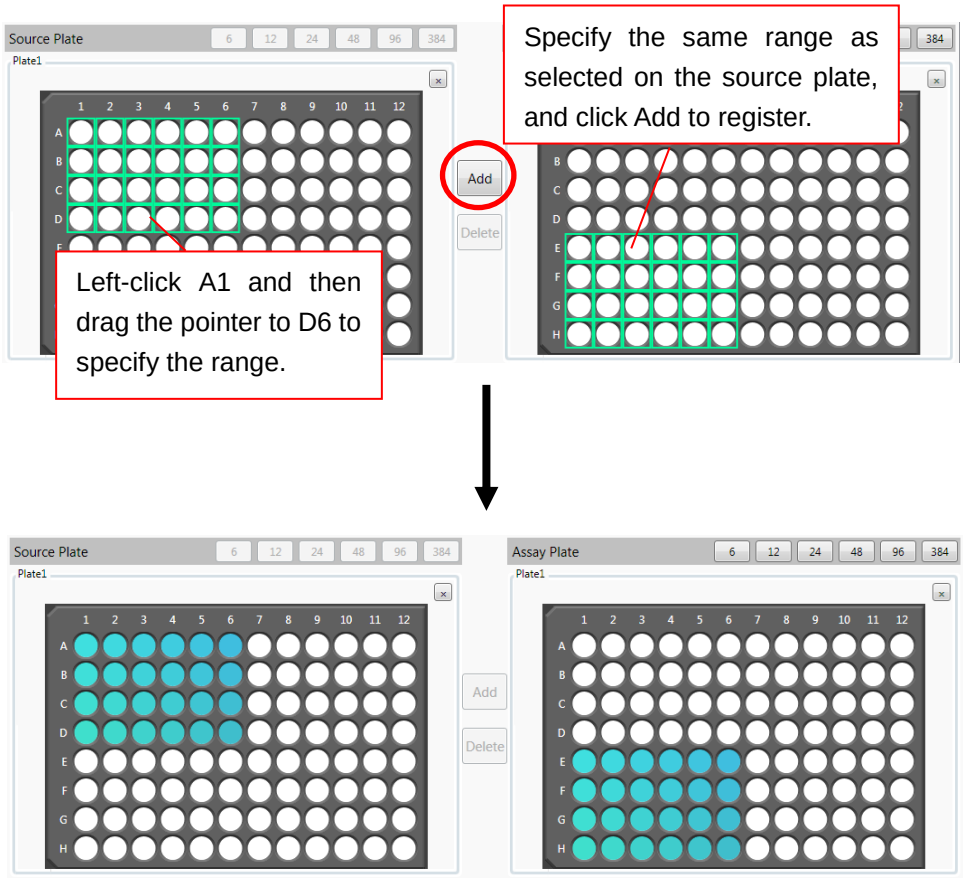
MEMO

- Dispensing to an assay plate from one well on the source plate is possible up to four wells.

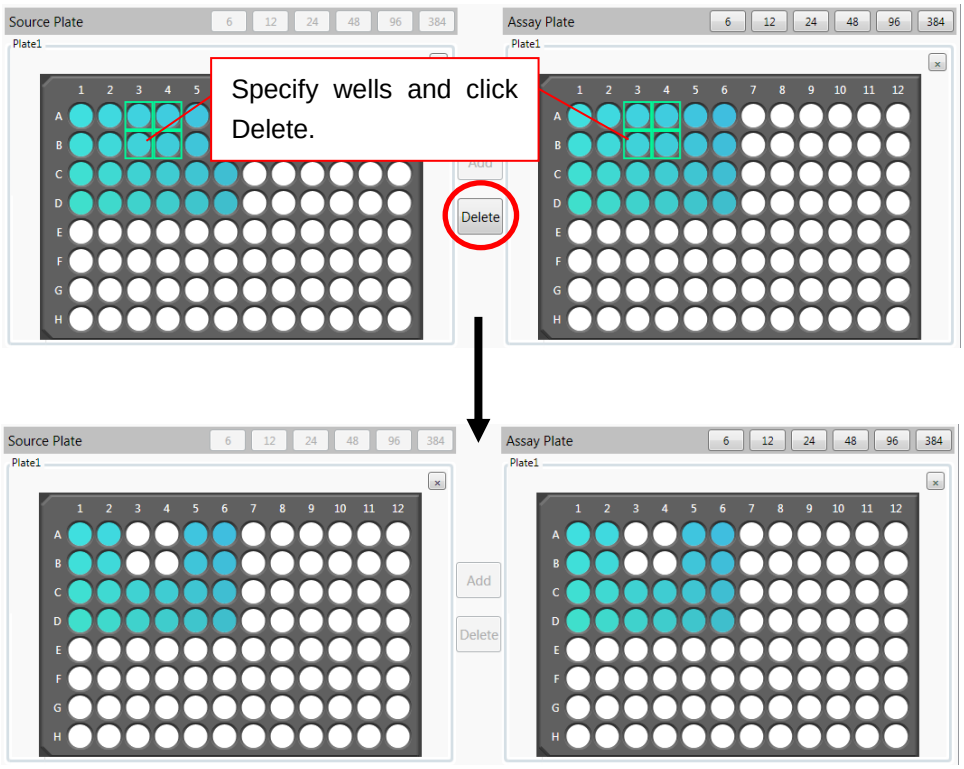
●When linking individual wells



●When linking wells by selecting the range



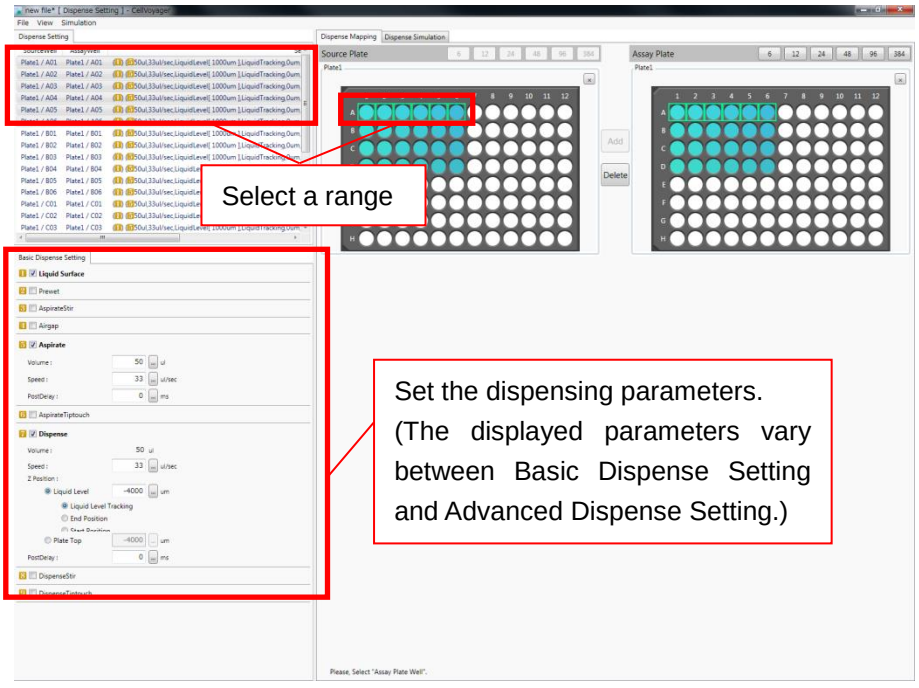
●When deleting wells



7) Set the dispensing parameters. (Refer to 5.13)

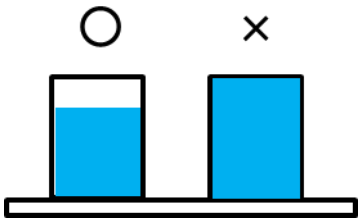
When the association is set, the setting items for individual wells are displayed on the “Dispense Setting” tab.

Select a range of wells in the source plate, or select a range of wells on the “Dispense Setting” tab by dragging with the mouse while holding down the Shift key, and then enter the dispensing parameters. The same parameters can be set to all wells in the selected range. The dispensing parameters to be entered vary between “Basic Dispense Setting” and “Advanced Dispense Setting.”



⚠ CAUTION

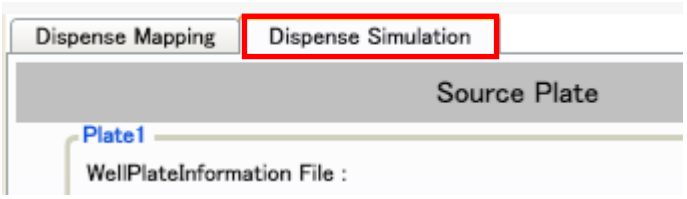
●Do NOT put solution up to each well top of Assay plate and Source plate.



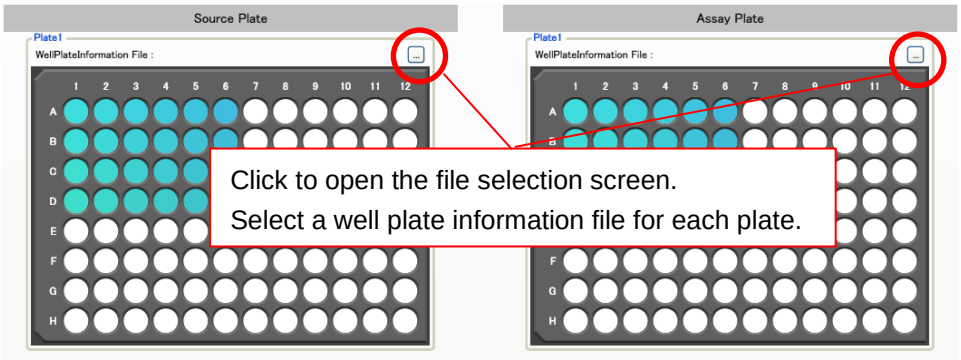
MEMO

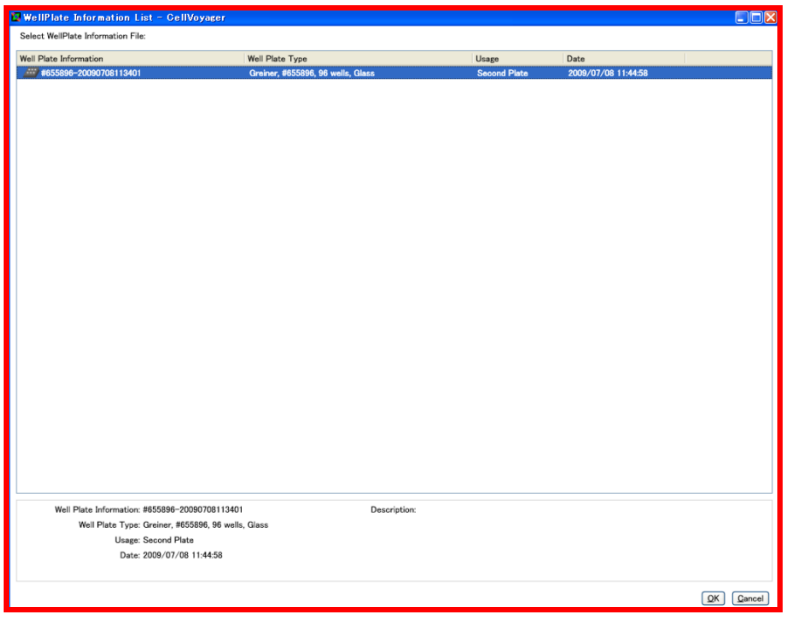
●Indication of reagent volume is 20% - 40% of well volume in Assay Plate and 50% - 80% of well volume in Source Plate.

8) Perform dispensing simulation.
Click the “Dispense Simulation” tab.



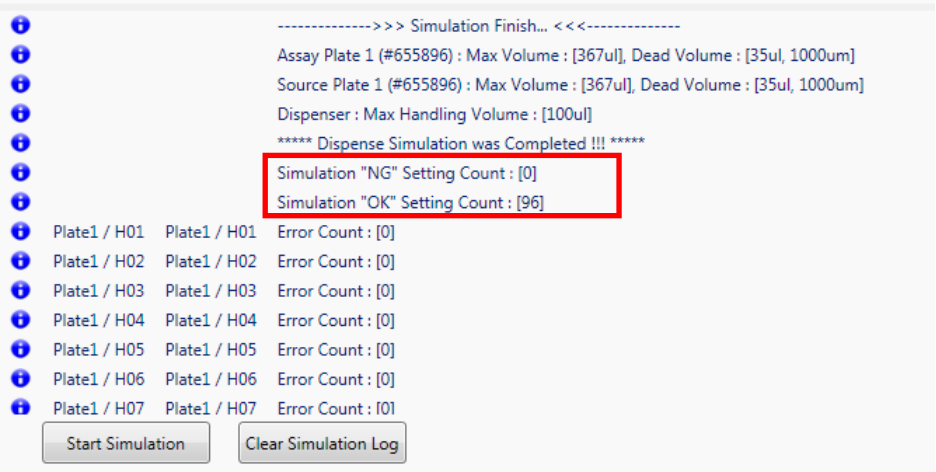
9) Select the well plate information file. (Refer to 4.1 for the creation of a well plate information file.)



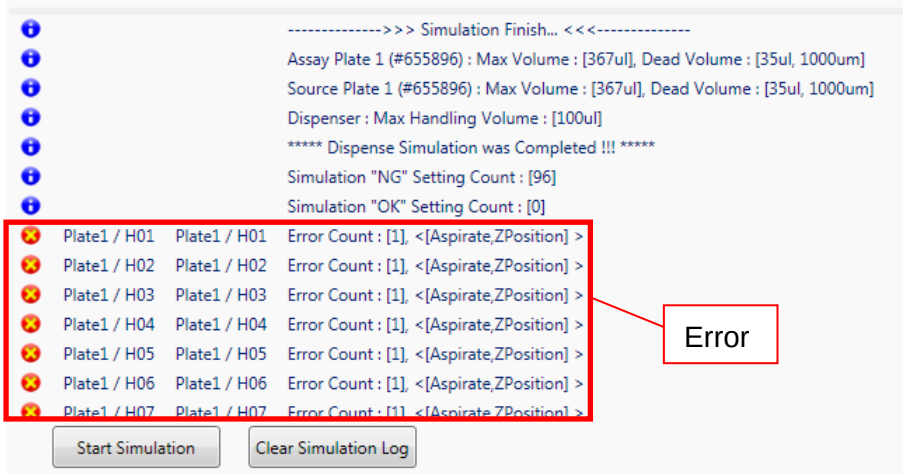


10) Click Start Simulation.

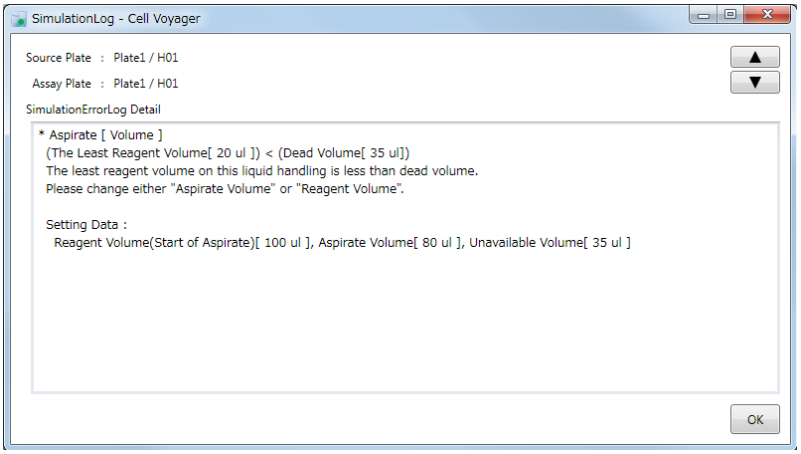
If the simulation is successful, the following message appears.



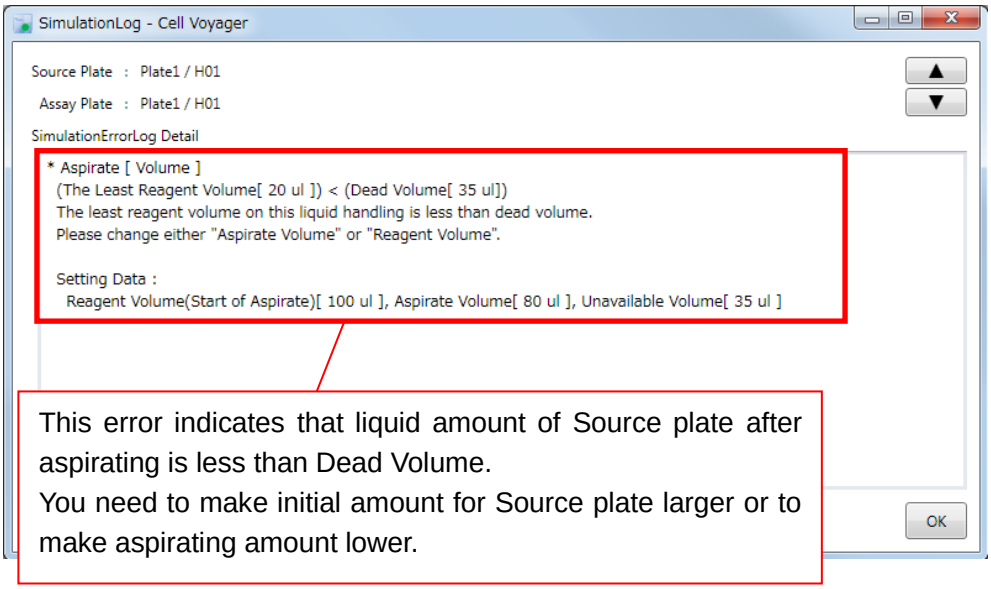
If an error occurs with the simulation, an error message appears. Change the error setting, and continue the simulation until an error message does not appear.



To confirm error in detail, double-click each of error items.

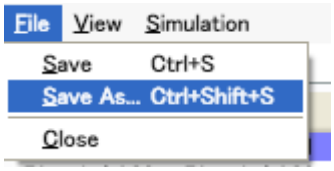


●Example of a simulation error



This error indicates that liquid amount of Source plate after aspirating is less than Dead Volume.
You need to make initial amount for Source plate larger or to make aspirating amount lower.

11) Save the dispensing setting file.
In the menu, click File -> Save As.



5.13. Setting the Dispensing Parameters

Basic Dispense Setting

MEMO

●Be sure to set Aspirate and Dispense.

Liquid Surface

This detects the liquid surface. It lowers the tip end in the well on the source plate so far that it comes in contact with the liquid surface to detect the liquid surface position in the well.
To set the Liquid Surface function, select the check box.

Prewet

This aspirates and drops the solution in the well on the source plate to make the inside of the tip wet with the solution.
To set the Prewet function, select the check box. Enter the value of “Volume" (amount of reagent suctioned).
(96-tip rack model: 0≦Volume≦100) (384-tip rack model: 0≦Volume≦20)

AspirateStir

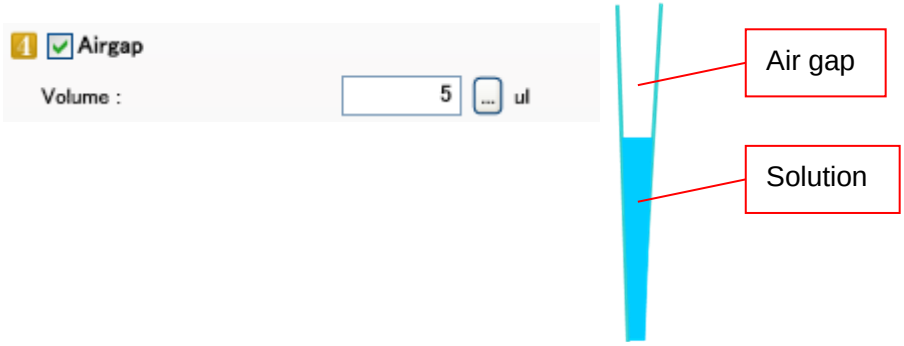
This stirs the solution in the well on the source plate. The solution is aspirated and discharged the number of times specified.
To set the AspirateStir function, select the check box. Enter the values of “Execution” (number of times solution is stirred) and “Volume" (amount of reagent suctioned). Also enter the value of “PostDelay” (sleep time after the syringe operation) if any high-viscosity solution is handled.
(1≦Execution≦5) (0≦PostDelay≦10000)
(96-tip rack model: 0≦Volume≦100) (384-tip rack model: 0≦Volume≦20)

Airgap

This delivers air on top of the solution inside the tip. It drops the solution to the last drop.

To set the Airgap function, select the check box. Enter the value of “Volume” (air gap).

(96-tip rack model: 0≦Volume≦100) (384-tip rack model: 0≦Volume≦20)



Aspirate

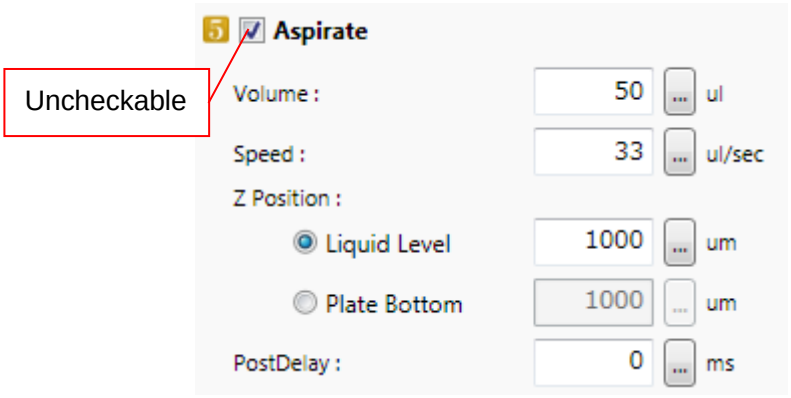
This aspirates the solution from the well on the source plate.

Enter the values of “Volume” (amount of reagent filled), “Speed” (filling speed) and “Z Position” (filling position). Also enter the value of “PostDelay” (sleep time after the syringe operation) if any high-viscosity solution is handled.

(96-tip rack model: 0≦Volume≦100) (384-tip rack model: 0≦Volume≦20)

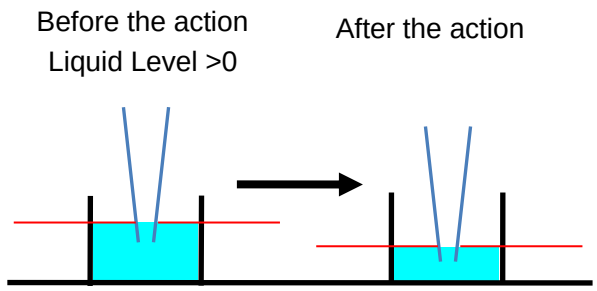
(96-tip rack model: 0≦Speed≦34) (384-tip rack model: 0≦Speed≦7)

(0≦PostDelay≦10000) (-50000≦LiquidLevel、PlateBottom≦100000)

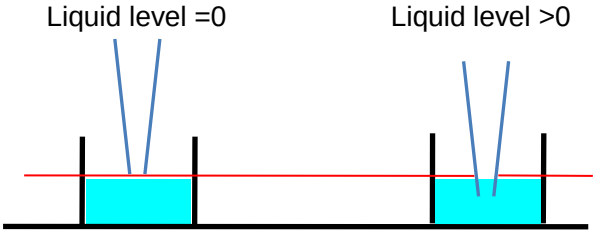


Z Position	Explanation
Liquid Level	Track the drop in liquid level due to filling. Input value indicates the distance between the tip and liquid level. (+ : Below the liquid level, 0 : Around the liquid level, - : Above the liquid level)
Plate Bottom	Fill the solution by using the bottom face of the plate as a reference. (+ : Below the bottom face of the plate, 0 : Around the bottom face of the plate, - : Above the bottom face of the plate)

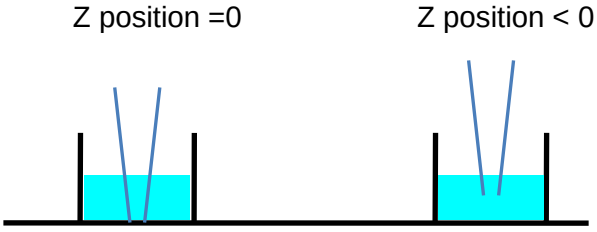
●Liquid Level tracks the fall of the liquid level that occurs as the solution is filled and lowers the tip position in accordance with the fall of the liquid level. The relative position of the liquid level and tip is fixed.



Liquid Level indicates the distance from the tip to the liquid surface. The plus indicates the downward tip position and the minus indicates the upward tip position in relation to the liquid level.



●With regards to Plate Bottom, the plus indicates the downward tip position and the minus indicates the upward tip position in relation to the bottom plane of the well. Enter minus to set.



AspirateTiptouch

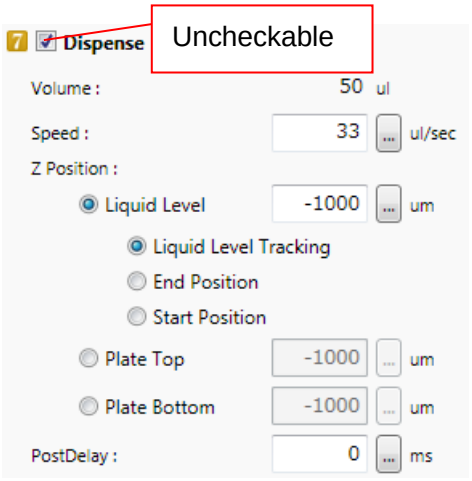
This lets the tip come in contact with the well wall on the source plate and drops the droplet adhered to the end.
To set the AspirateTiptouch function, select the check box.

Dispense

This enables you to set how to drop the solution.

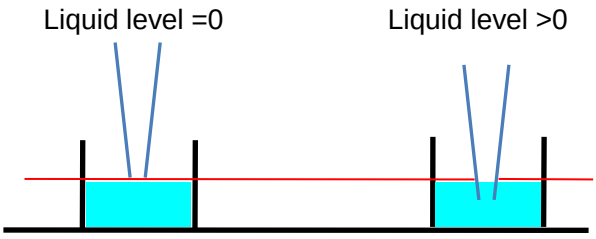
Enter the values of “Speed” (dripping speed) and “Z Position” (dripping position). Also enter the value of “PostDelay” (sleep time after the syringe operation) if any high-viscosity solution is handled. The value of “Volume” (amount of solution dripped into each well) is calculated automatically based on the set parameters.

(96-tip rack model: 0≦Speed≦34) (384-tip rack model: 0≦Speed≦7)
(-50000≦LiquidLevel, PlateTop, PlateBottom≦100000)
(0≦PostDelay≦10000)

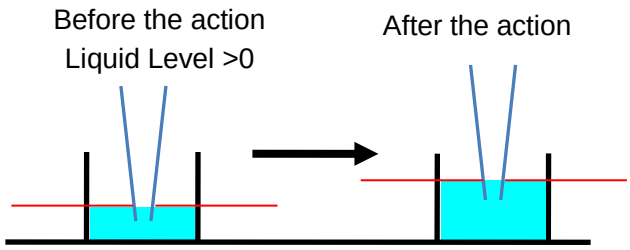


Z Position	Explanation
Liquid Level	Distance between the tip and liquid level (+ : Below the liquid level, 0 : Around the liquid level, - : Above the liquid level)
Liquid Level Tracking	Track the rise in liquid level due to dripping.
End Position	Drip the solution by using the liquid level after dripping as a reference.
Start Position	Drip the solution by using the liquid level before dripping as a reference.
Plate Top	Drop by using the top face of the plate as a reference. (+ : Below the top face of the plate, 0 : Around the top face of the plate, - : Above the top face of the plate)
Plate Bottom	Drop by using the bottom face of the plate as a reference. (+ : Below the bottom face of the plate, 0 : Around the bottom face of the plate, - : Above the bottom face of the plate)

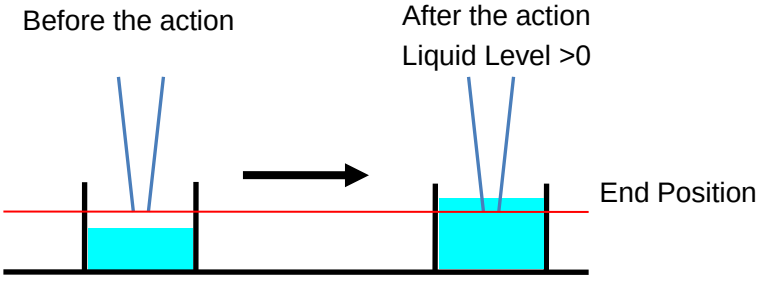
●Liquid Level indicates the distance from the tip to the liquid level. The plus indicates the downward tip position and the minus indicates the upward tip position in relation to the liquid level.



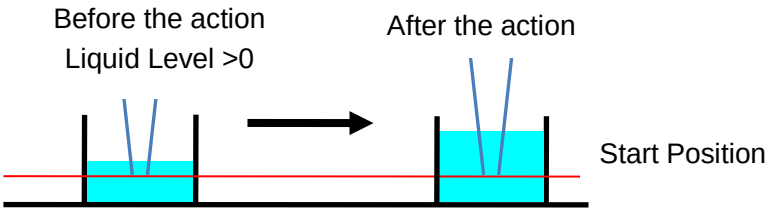
●Liquid Level Tracking tracks the rise of the liquid level that occurs as the solution is dropped and raises the tip position in accordance with the rise of the liquid level. The relative position of the liquid level and tip is fixed.



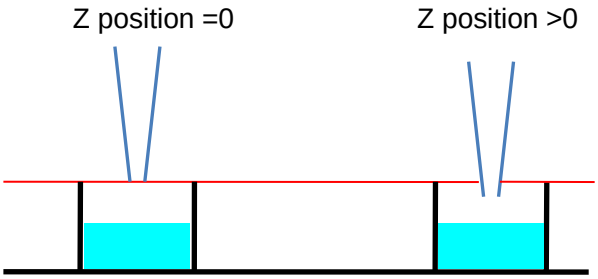
●End Position sets the tip to a position that is shifted from the liquid level after the action by the liquid level you enter. The tip position before and after the action is unchanged.



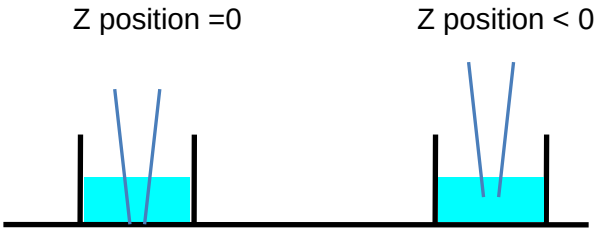
●Start Position sets the tip to a position that is shifted from the liquid level before the action by the liquid level you enter. The tip position before and after the action is unchanged.



●With regards to Plate Top, the plus indicates the downward tip position and the minus indicates the upward tip position in relation to the top plane of the well.



●With regards to Plate Bottom, the plus indicates the downward tip position and the minus indicates the upward tip position in relation to the bottom plane of the well. Enter minus to set.



DispenseStir

This stirs the solution in the well after the solution is dropped. The solution in the well is aspirated and discharged the specified number of times.

To set the DispenseStir function, select the check box. Enter the values of “Execution” (number of times solution is stirred) and “Volume" (amount of reagent suctioned). Also enter the value of “PostDelay” (sleep time after the syringe operation) if any high-viscosity solution is handled.

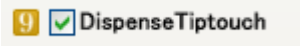
(1≦Execution≦5) (0≦PostDelay≦10000)

(96-tip rack model: 0≦Volume≦100) (384-tip rack model: 0≦Volume≦20)

DispenseTiptouch

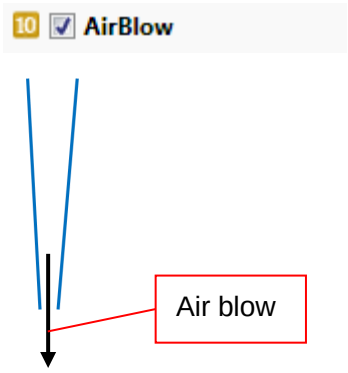
This lets the tip come in contact with the well wall on the assay plate and drops the droplet adhered to the end.

To set the DispenseTiptouch function, select the check box.



AirBlow

This allows to blow air from inside tip to drop droplet adherent on the tip top. To set the AirBlow function, select the check box.
This function can be used only when liquid amount (including Airgap) inside tip is 0ul.



Advanced Dispense Setting

MEMO

●Be sure to set Aspirate and Dispense.

Liquid Surface

This detects the liquid surface. It lowers the tip end in the well on the source plate so far that it comes in contact with the liquid surface and detects the liquid surface position in the well.
To set the Liquid Surface function, select the check box.

Prewet

This aspirates and drops the solution in the well on the source plate to make the inside of the tip wet with the solution.
To set the Prewet function, select the check box. Enter the value of “Volume" (amount of reagent suctioned).
(96-tip rack model: 0≦Volume≦100) (384-tip rack model: 0≦Volume≦20)

AspirateStir

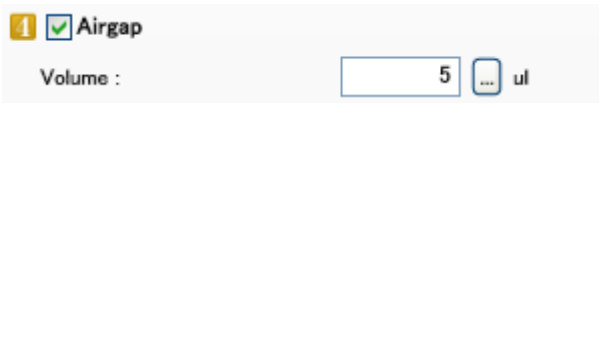
This stirs the solution in the well on the source plate. The solution is aspirated and discharged the specified number of times.
To set the AspirateStir function, select the check box. Enter the values of “Execution” (number of times solution is stirred) and “Volume" (amount of reagent suctioned). Also enter the value of “PostDelay” (sleep time after the syringe operation) if any high-viscosity solution is handled.
(1≦Execution≦5) (0≦PostDelay≦10000)
(96-tip rack model: 0≦Volume≦100) (384-tip rack model: 0≦Volume≦20)

Airgap

This delivers air on top of the solution inside the tip to drop the solution to the last drop.

To set the Airgap function, select the check box. Enter the value of “Volume” (air gap).

(96-tip rack model: 0≦Volume≦100) (384-tip rack model: 0≦Volume≦20)



Aspirate

This aspirates the solution from the well on the source plate.

Enter the values of “Volume” (amount of reagent filled), “Speed" (filling speed) and “Z Position” (filling position). Also enter the values of “X Position” and “Y Position” if the tip position at the time of filling is to be specified. Enter the values of “PreDelay” (sleep time before the syringe operation) and “PostDelay” (sleep time after the syringe operation) if any high-viscosity solution is handled.

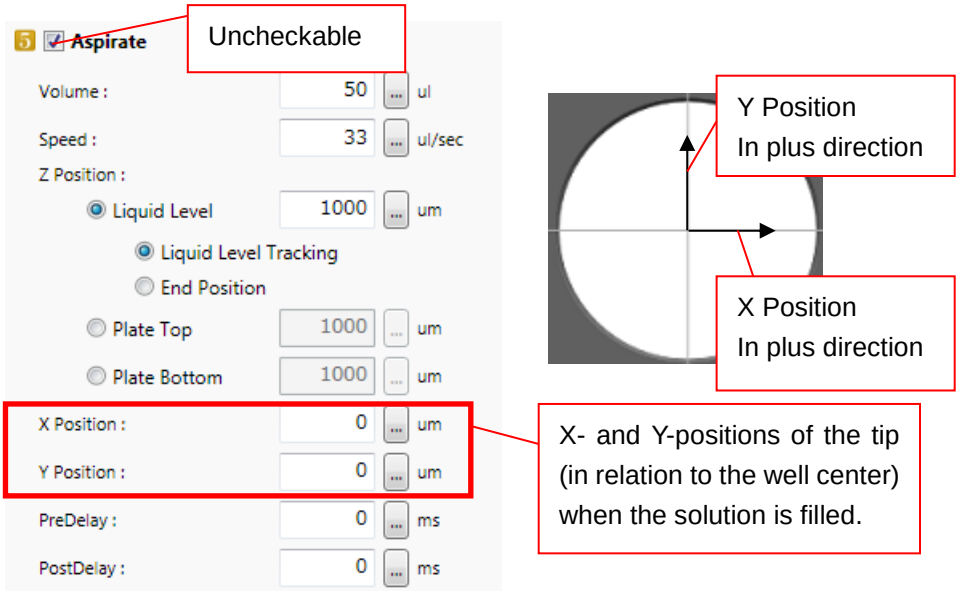
(96-tip rack model: 0≦Volume≦100) (384-tip rack model: 0≦Volume≦20)

(96-tip rack model: 0≦Speed≦34) (384-tip rack model: 0≦Speed≦7)

(-50000≦LiquidLevel, PlateTop, PlateBottom≦100000)

(-50000≦X Position, Y Position≦50000)

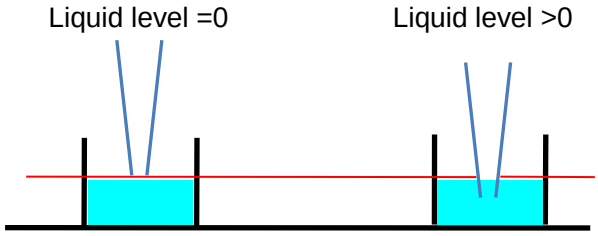
(0≦PreDelay, PostDelay≦10000)



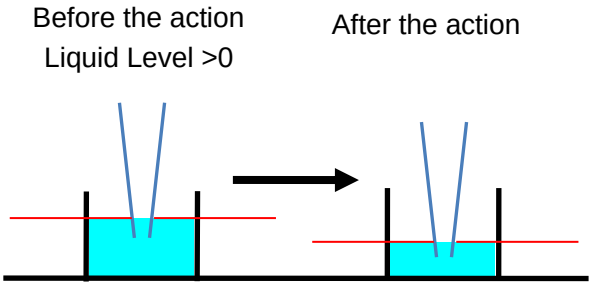
Z Position	Explanation
Liquid Level	Distance between the tip and liquid level (+ : Below the liquid level, 0 : Around the liquid level, - : Above the liquid level)
Liquid Level Tracking	Track the drop in liquid level due to filling.

End Position	Fill the solution by using the liquid level after filling as a reference.
Plate Top	Fill the solution by using the top face of the plate as a reference. (+ : Below the top face of the plate, 0 : Around the top face of the plate, - : Above the top face of the plate)
Plate Bottom	Fill the solution by using the bottom face of the plate as a reference. (+ : Below the bottom face of the plate, 0 : Around the bottom face of the plate, - : Above the bottom face of the plate)

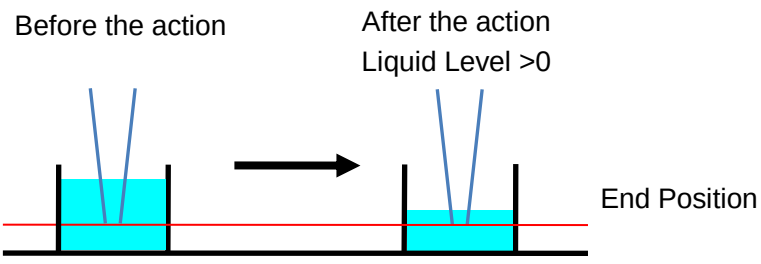
●Liquid Level indicates the distance from the tip to the liquid surface. The plus indicates the downward tip position and the minus indicates the upward tip position in relation to the liquid level.



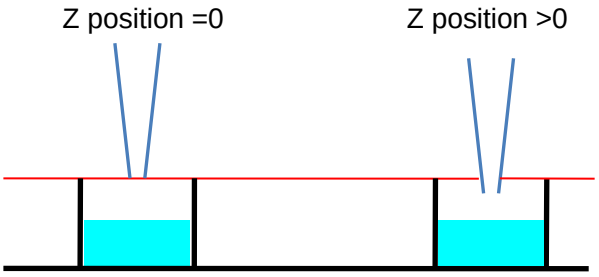
●Liquid Level Tracking tracks the fall of the liquid level that occurs as the solution is filled and lowers the tip position in accordance with the fall of the liquid level. The relative position of the liquid level and tip is fixed.



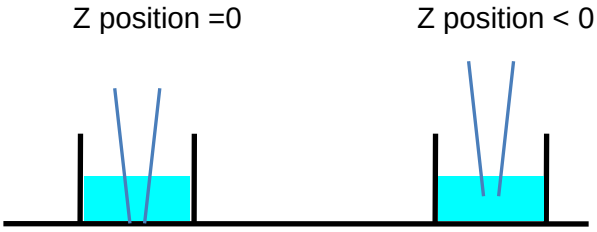
●End Position sets the tip to a position that is shifted from the liquid level after the action by the entered liquid level.



●With regards to Plate Top, the plus indicates the downward tip position and the minus indicates the upward tip position in relation to the well top plane.



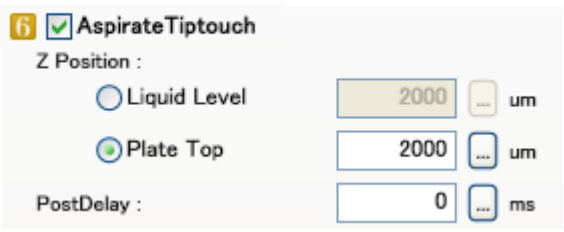
●With regards to Plate Bottom, the plus indicates the downward tip position and the minus indicates the upward tip position in relation to the bottom plane of the well. Enter minus to set.



AspirateTiptouch

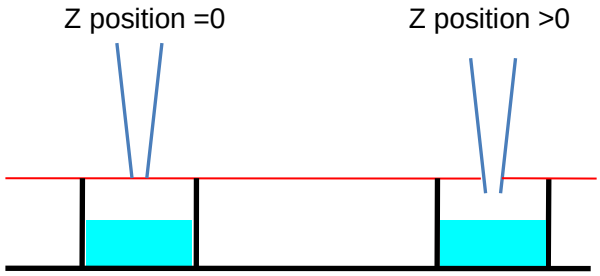
This lets the tip come in contact with the well wall and drops the droplet adhered to the end.

To set the AspirateTiptouch function, select the check box. Enter the value of “Z Position” (position at which the tip touches). Also enter the value of “PostDelay” (sleep time after the syringe operation) if any high-viscosity solution is handled. (0≦PlateTop≦2000) (0≦PostDelay≦10000)



Z Position	Explanation
Plate Top	Distance between the tip and top face of the plate (+ : Below the top face of the plate, 0 : Around the top face of the plate, - : Above the top face of the plate)

●With regards to Plate Top, the plus indicates the downward tip position and the minus indicates the upward tip position in relation to the well top plane.



Dispense

This sets how to drop the reagent.

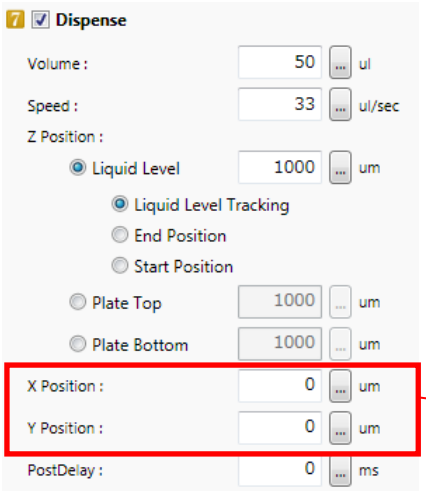
Enter the values of “Volume” (amount of reagent dripped into each well), “Speed” (dripping speed) and “Z Position” (dripping position). Also enter the values of “X Position” and “Y Position” if the tip position at the time of dripping is to be specified. Enter the value of “PostDelay” (sleep time after the syringe operation) if any high-viscosity solution is handled.

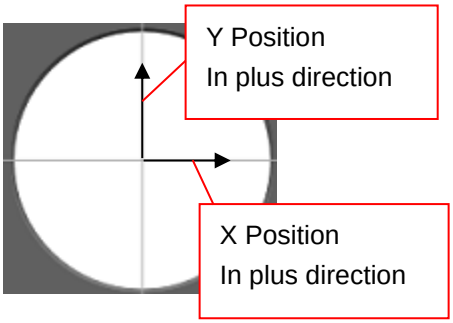
(96-tip rack model: 0≦Volume≦100) (384-tip rack model: 0≦Volume≦20)

(96-tip rack model: 0≦Speed≦34) (384-tip rack model: 0≦Speed≦7)

(-50000≦LiquidLevel, PlateTop, PlateBottom≦100000)

(-50000≦X Position, Y Position≦50000) (0≦PostDelay≦10000)



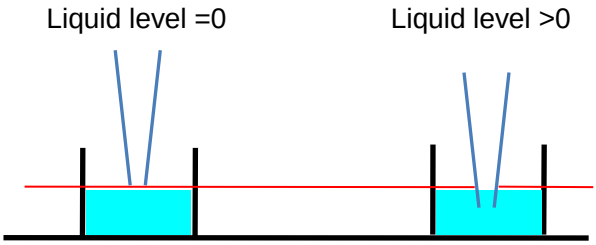


X- and Y-positions when the reagent is dropped (in relation to the well center)

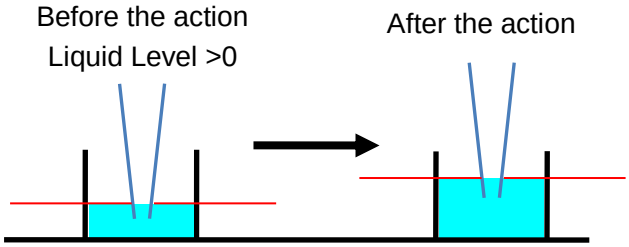
Z Position	Explanation
Liquid Level	Distance between the tip and liquid level (+ : Below the liquid level, 0 : Around the liquid level, - : Above the liquid level)
Liquid Level Tracking	Track the rise in liquid level due to dripping.
End Position	Drip the solution by using the liquid level after dripping as a reference.
Start Position	Drip the solution by using the liquid level before dripping as a reference.
Plate Top	Drop by using the top face of the plate as a reference. (+ : Below the top face of the plate, 0 : Around the top face of the plate, - : Above the top face of the plate)

Plate Bottom	Drop by using the bottom face of the plate as a reference. (+ : Below the bottom face of the plate, 0 : Around the bottom face of the plate, - : Above the bottom face of the plate)

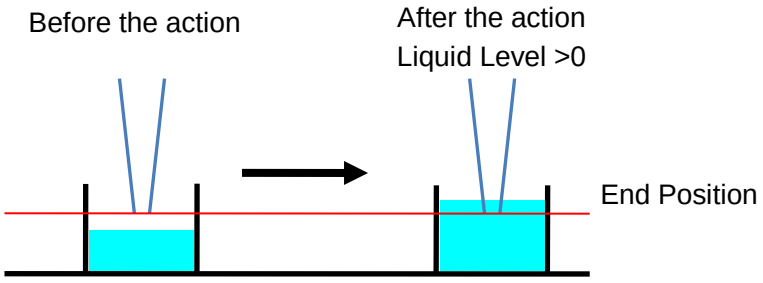
●Liquid Level indicates the distance from the tip to the liquid surface. The plus indicates the downward tip position and the minus indicates the upward tip position in relation to the liquid level.



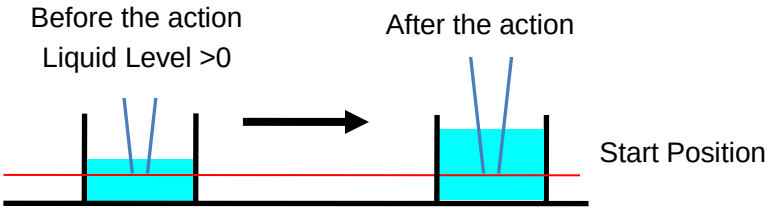
●Liquid Level Tracking tracks the fall of the liquid level that occurs as the solution is filled and lowers the tip position in accordance with the fall of the liquid level. The relative position of the liquid level and tip is fixed.



●End Position sets the tip to a position that is shifted from the liquid level after the action by the entered liquid level.
The tip position does not change before and after the action.

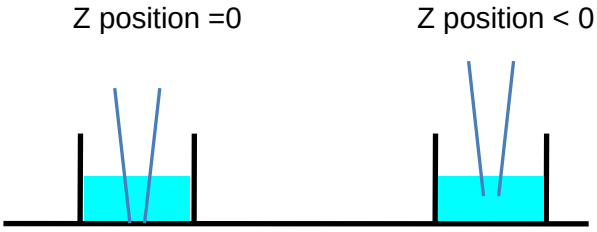


●Start Position sets the tip to a position that is shifted from the liquid level before the action by the liquid level you enter.
The tip position does not change before and after the action.



●With regards to Plate Top, the plus indicates the downward tip position and the minus indicates the upward tip position in relation to the top plane of the well.

●With regards to Plate Bottom, the plus indicates the downward tip position and the minus indicates the upward tip position in relation to the bottom plane of the well. Enter minus to set.



DispenseStir

This stirs the dropped solution in the well. The solution in the well is aspirated and discharged the specified number of times.

To set the DispenseStir function, select the check box. Enter the values of “Execution” (number of times solution is stirred) and “Volume" (amount of reagent suctioned). Also enter the value of “PostDelay” (sleep time after the syringe operation) if any high-viscosity solution is handled.

(1≦Execution≦5) (0≦PostDelay≦10000)

(96-tip rack model: 0≦Volume≦100) (384-tip rack model: 0≦Volume≦20)

DispenseTiptouch

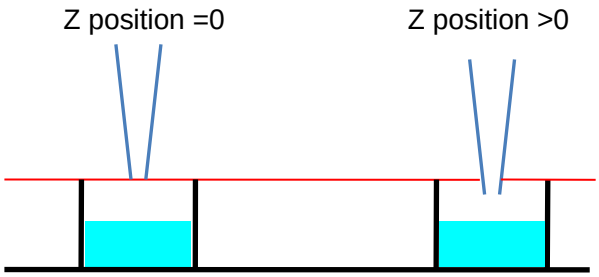
This lets the tip come in contact with the well wall to drop the solution adhered to the end.

To set the DispenseTiptouch function, select the check box. Enter the value of “Z Position” (position at which the tip touches). Also enter the value of “PostDelay” (sleep time after the syringe operation) if any high-viscosity solution is handled.

(0≦PlateTop≦2000) (0≦PostDelay≦10000)

Z Position	Explanation
Plate Top	Distance between the tip and top face of the plate (+ : Below the top face of the plate, 0 : Around the top face of the plate, - : Above the top face of the plate)

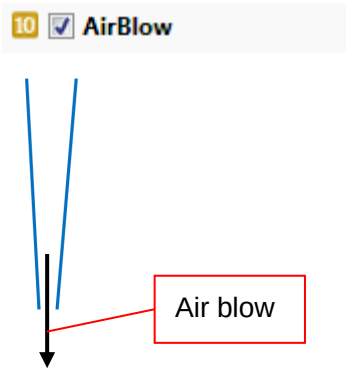
●With regards to Plate Top, the plus indicates the downward tip position and the minus indicates the upward tip position in relation to the well top plane.



AirBlow

This allows to blow air from inside tip to drop droplet adherent on the tip top. To set the AirBlow function, select the check box.

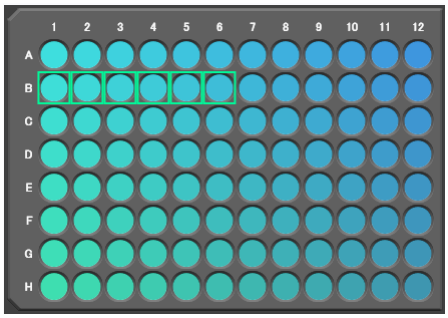
This function can be used only when liquid amount (including Airgap) inside tip is 0ul.



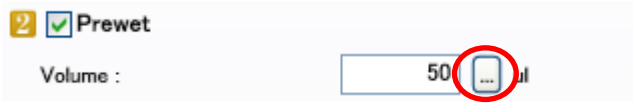
Auxiliary Input Function

This function is useful when entering the dispensing parameters for the wells in the specified range by changing their values at an equal distance, etc.

1) Specify a range of wells.

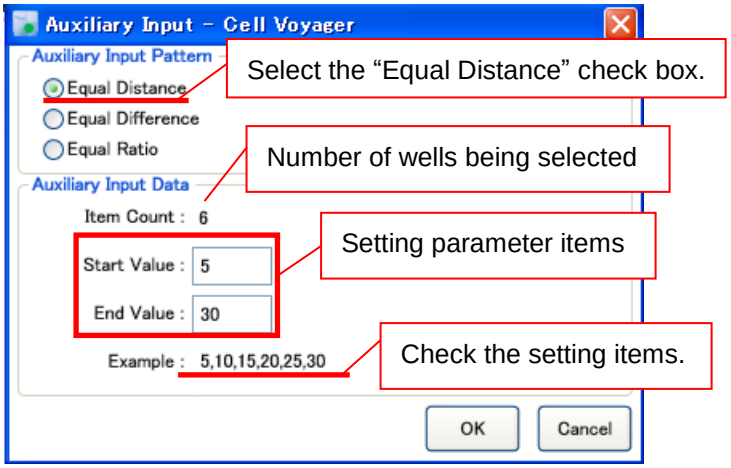


2) Click the button next to the entry box to open the auxiliary input screen.



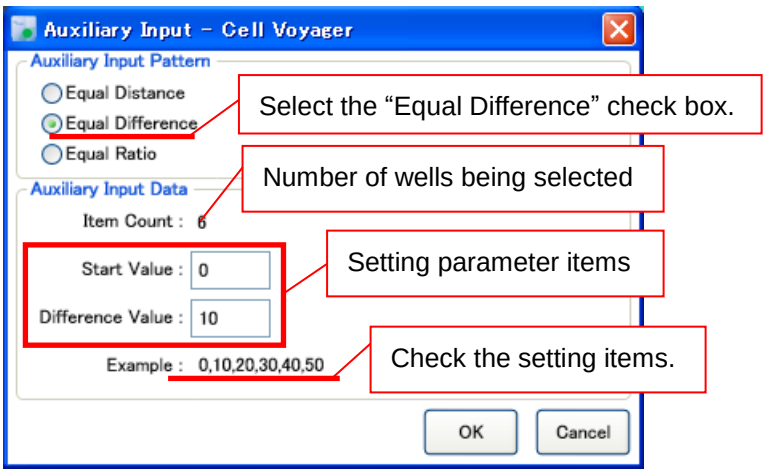
Equal Distance

Values are entered for the wells in the specified range at an equal distance. Enter the values of “Start Value” (value entered for the first well) and “End Value” (value entered for the last well).



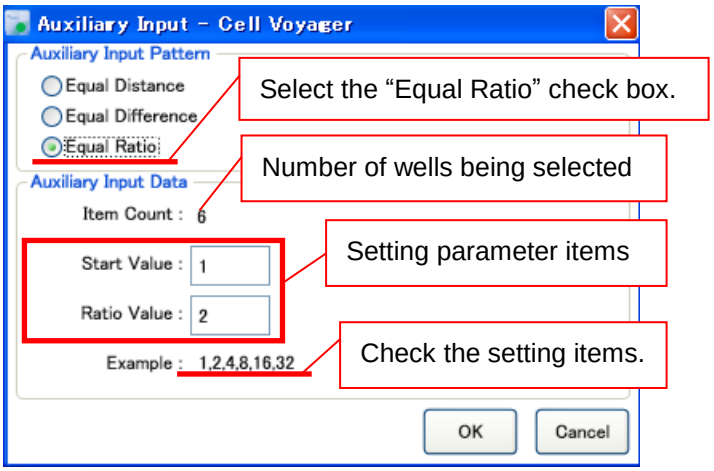
Equal Difference

Values are entered for the wells in the specified range at an equal difference. Enter the values of “Start Value” (value entered for the first well) and “Difference Value” (value of equal difference).



Equal Ratio

Values are entered for the wells in the specified range at an equal ratio. Enter the values of “Start Value” (value entered for the first well) and “Ratio Value” (value of equal ratio).



5.14. Setting the Water Immersion Lenses

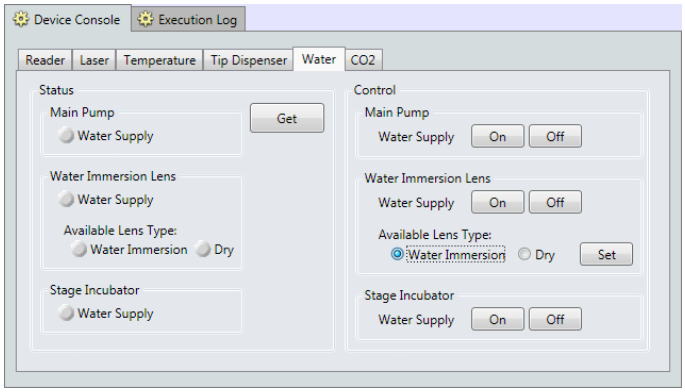
This setting is necessary to use water immersion lenses.
(Water immersion lens model only)

1) Prepare the water supply bottle and the drainage bottle. Put pure water into the water supply bottle. (Pour off the water in the bottles regularly.)

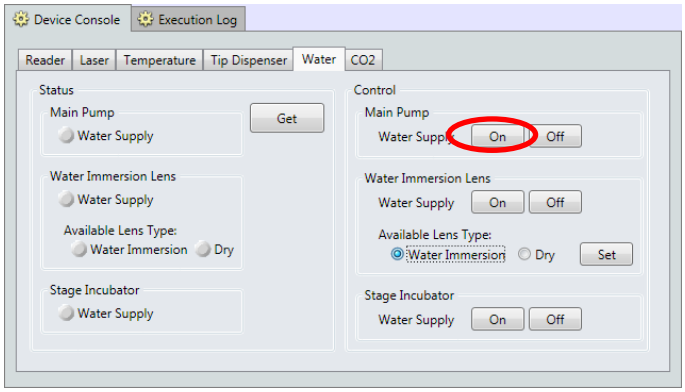
WARNING

- In the case of replacing water in the bottles, make sure that the MAIN POWER breaker surely turns OFF to shut down CV7000. (Refer to 3.5) Be careful not to turn the breaker ON by oversight while at work.

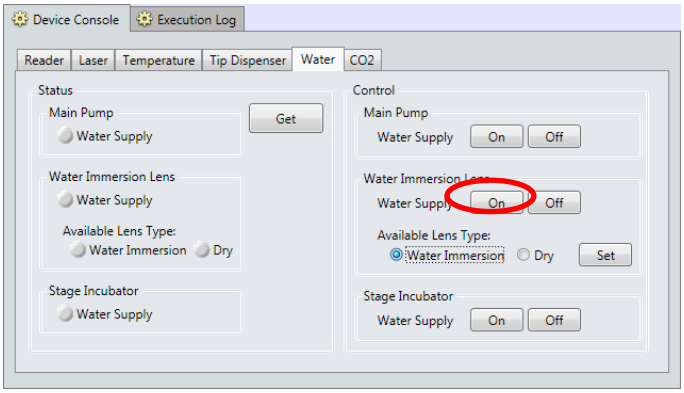
2) Open the “Water” tab within the “Device Console” tab (refer to 6.4) on the “Reader Control” screen.



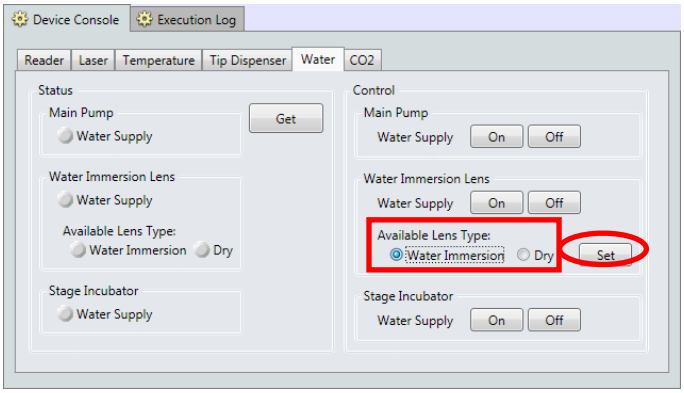
3) To supply the water in the main pump, click “On” of “Water Supply” in the “Main Pump” area. Wait for about ten minutes after clicking.



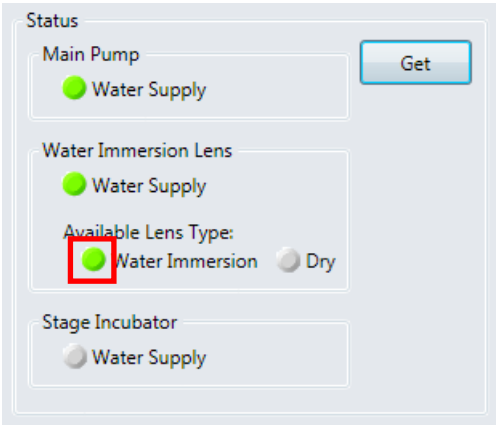
4) Click “On” of “Water Supply” in the “Water Immersion Lens” area.



5) Select “Water Immersion” and click “Set” If you use the water immersion lenses. Select “Dry” and click “Set” if you use the dry lenses.

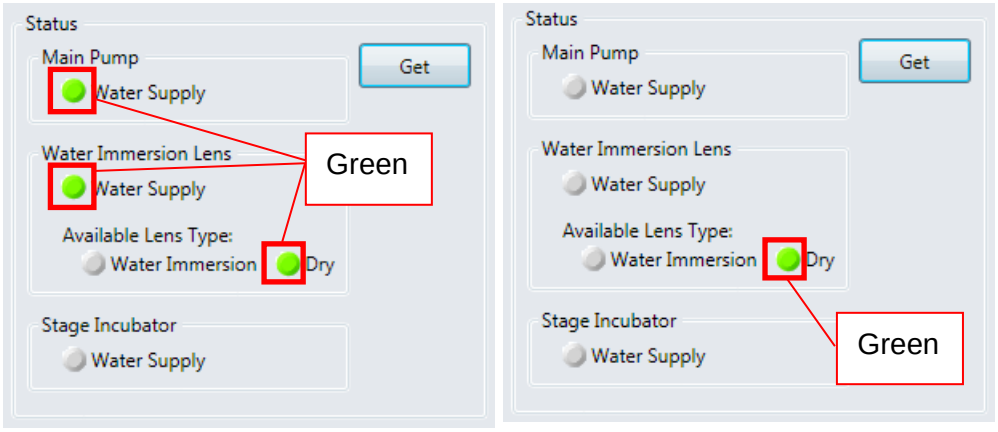


6) Click “Get” and check the current status information. Confirm the illuminating green icon of “Water Immersion”.

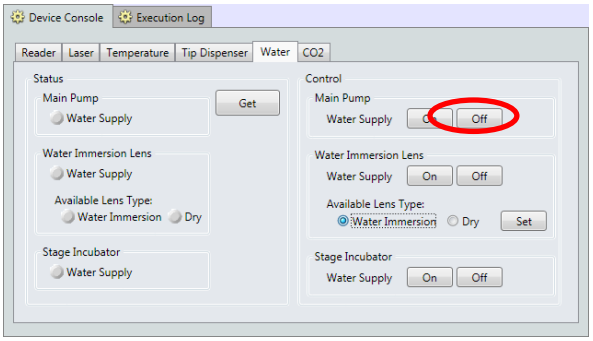


MEMO

●You can use the dry lenses in the examples below.



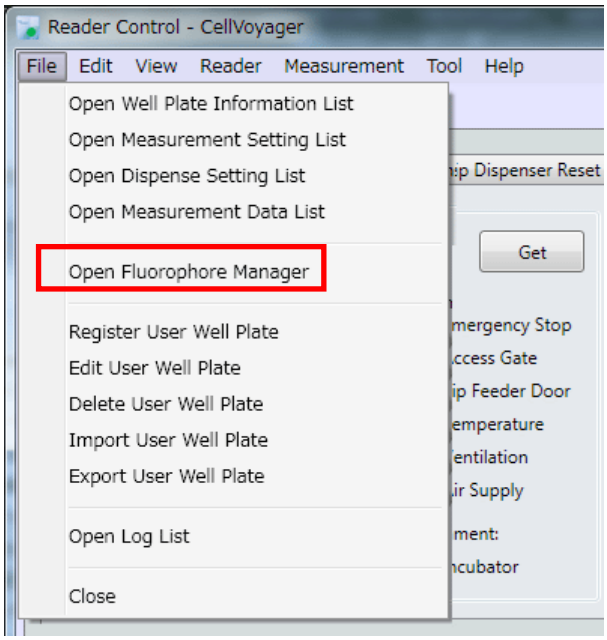
7) Click “Off” of “Water Supply” in the “Main Pump” area before the equipment is turned off. The water is drained away from the main pump.



5.15. Registering Fluorophore

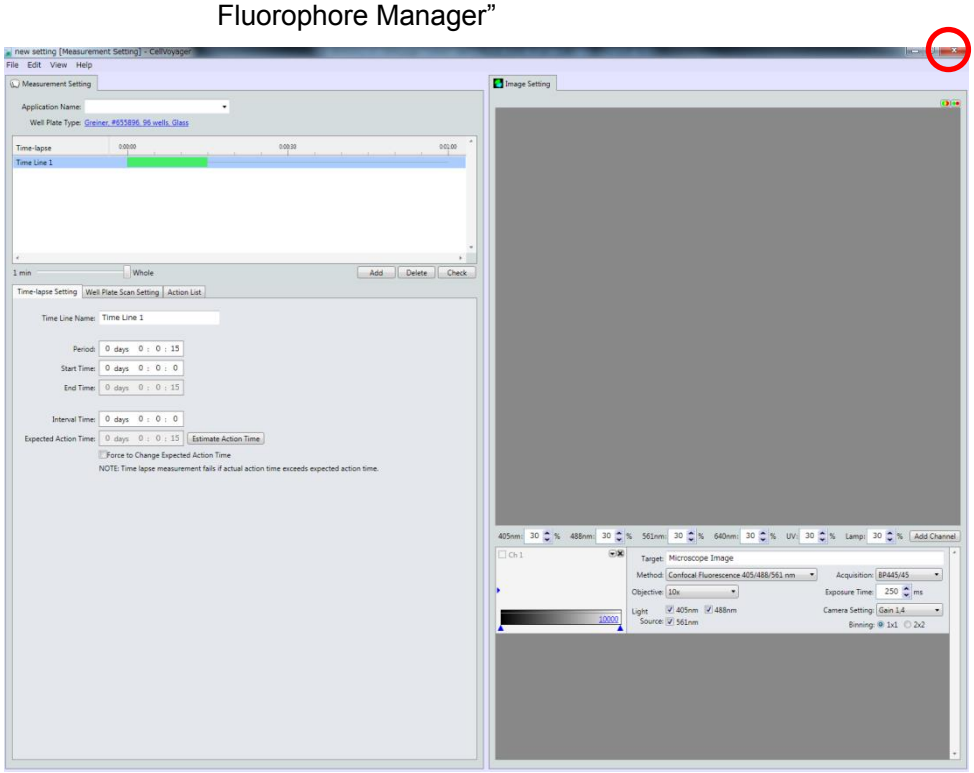
Register spectral information of fluorophore which is used to perform crosstalk correction by Image Correction Software

1) Select “File” -> “Open Fluorophore Manager” from menu of “Reader Control” window.

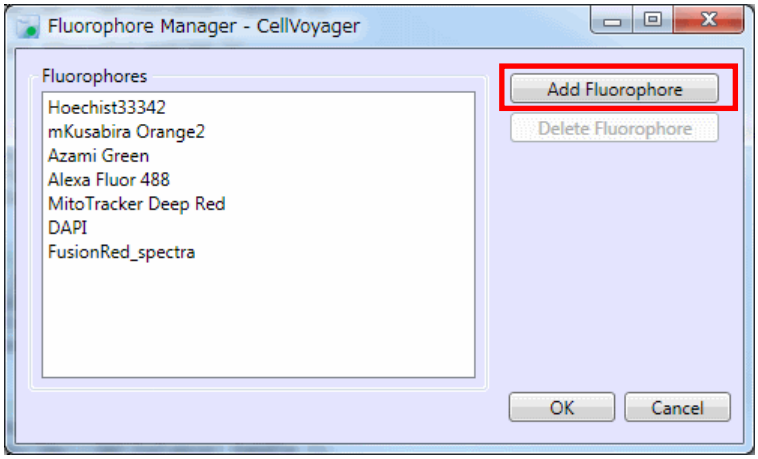


MEMO

●”Open Fluorophore Manager” cannot be selected when “Measurement Setting” window is open.
Close ”Measurement Setting” window and select “Open Fluorophore Manager”



2) Click “Add Fluorophore”



3) Load fluorophore spectral data file.

MEMO

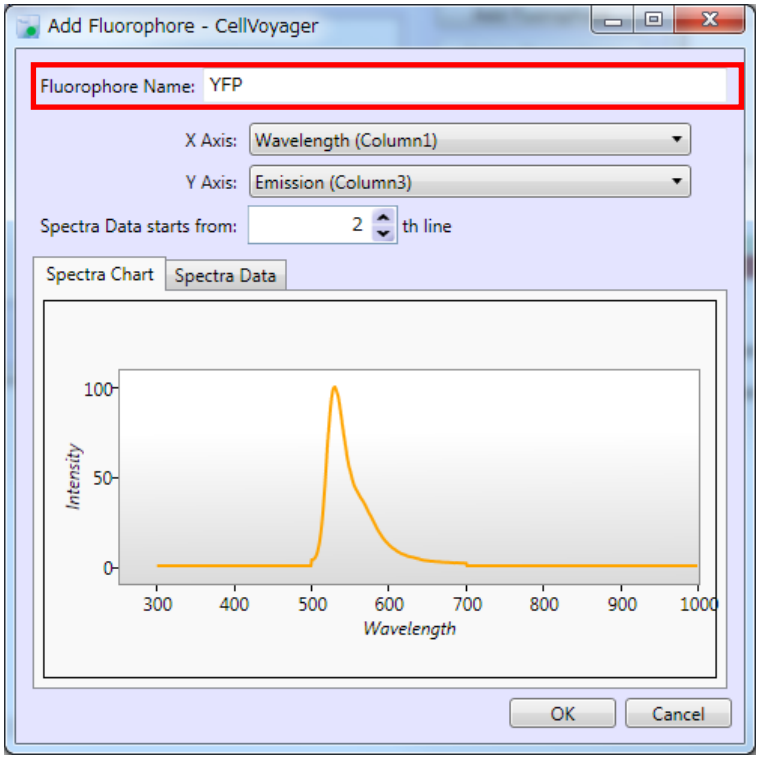
● Fluorephore spectral data file must meet following condition.

- CSV format
- It contains wavelength and emission spectral data.
- Spectral data is normalized maximum value to be 100.
- Data is arranged in the column direction as following. Wavelength and spectral data can be started any column.

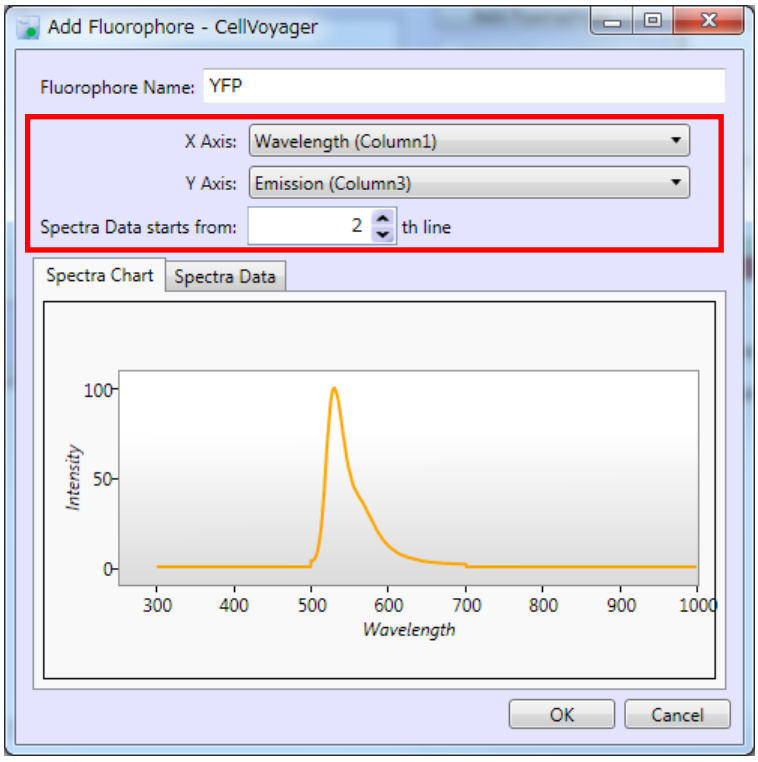
Wavelength	Emission
300	0
301	0
302	0
⋮	⋮

●Fluorephore spectral data file may be divided to excitation and emission. Load emission spectral data file.

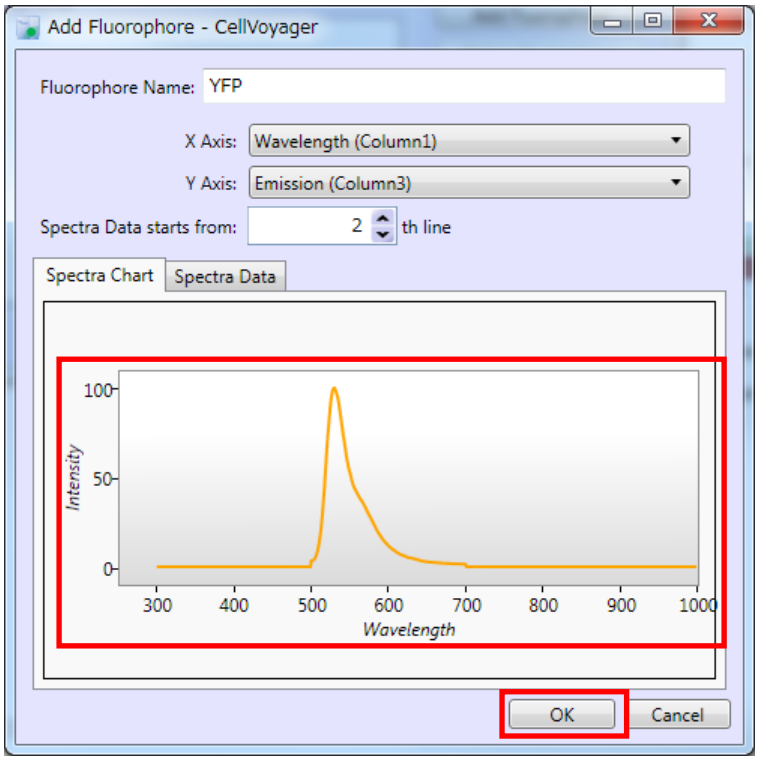
4) Fluorophore spectral data file name is input in “Fluorophore Name” automatically. Change it as needed.



5) Select wavelength data column as “X Axis” and emission spectral data column as “Y Axis”.
Input “Spectra Data starts from * th line”.

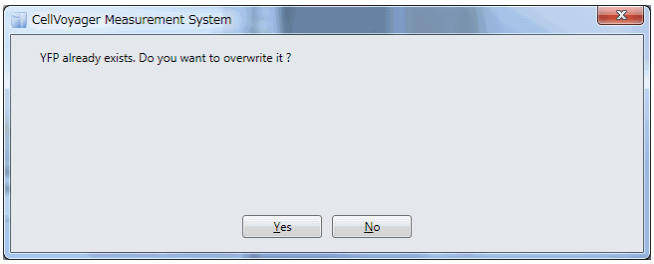


6) Confirm if there are no problems in emission spectral data and click “OK”.

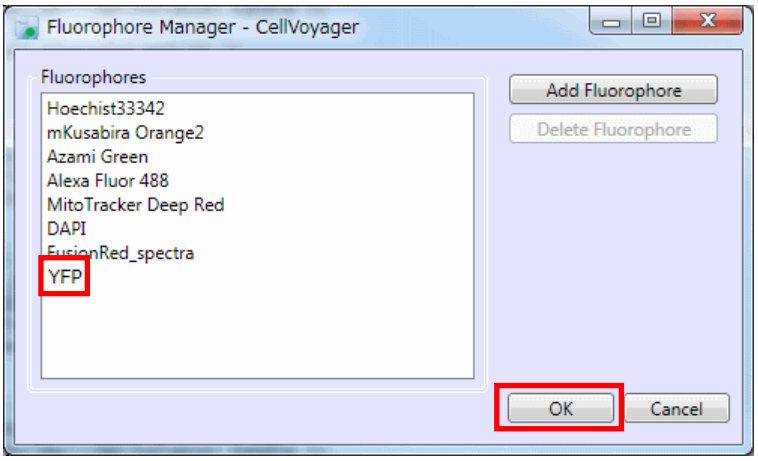


MEMO

● Following dialog is appeared if “Fluorophore Name” is same as that of already registered. Click “Yes” to overwrite emission spectral data. Click “No” to return to “Add Fluorophore” window.



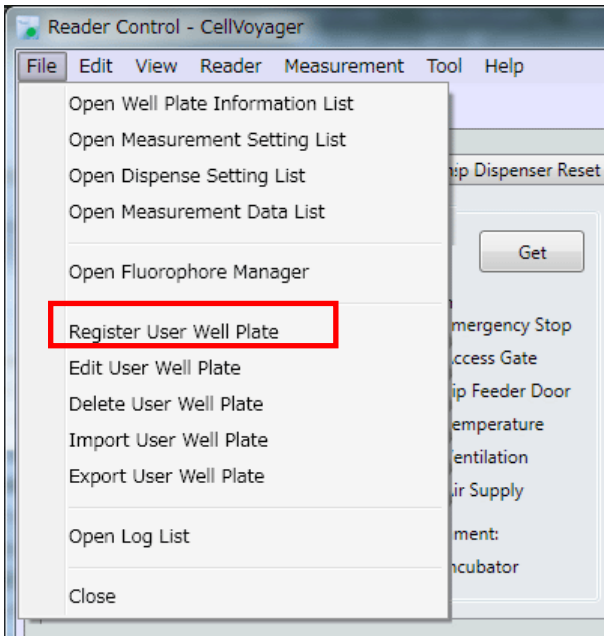
7) New fluorophore is added in “Fluorophores” list. Click “OK”.



5.16. Registering Well Plate

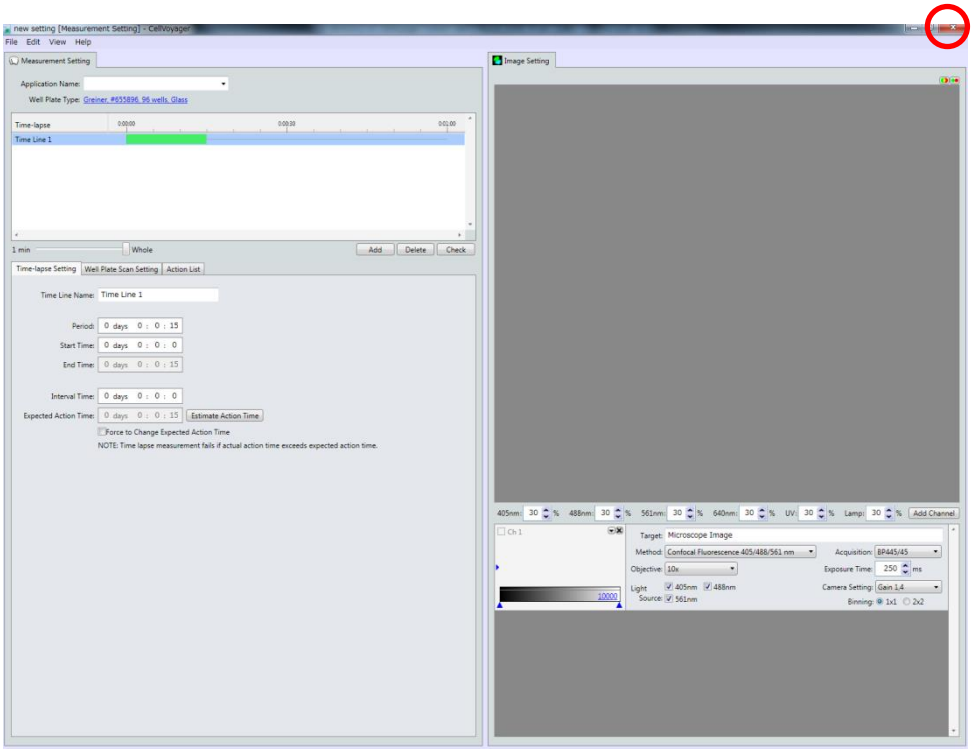
Register User Well Plate

1) Select “File” -> “Register User Well Plate” from menu of “Reader Control” window.



MEMO

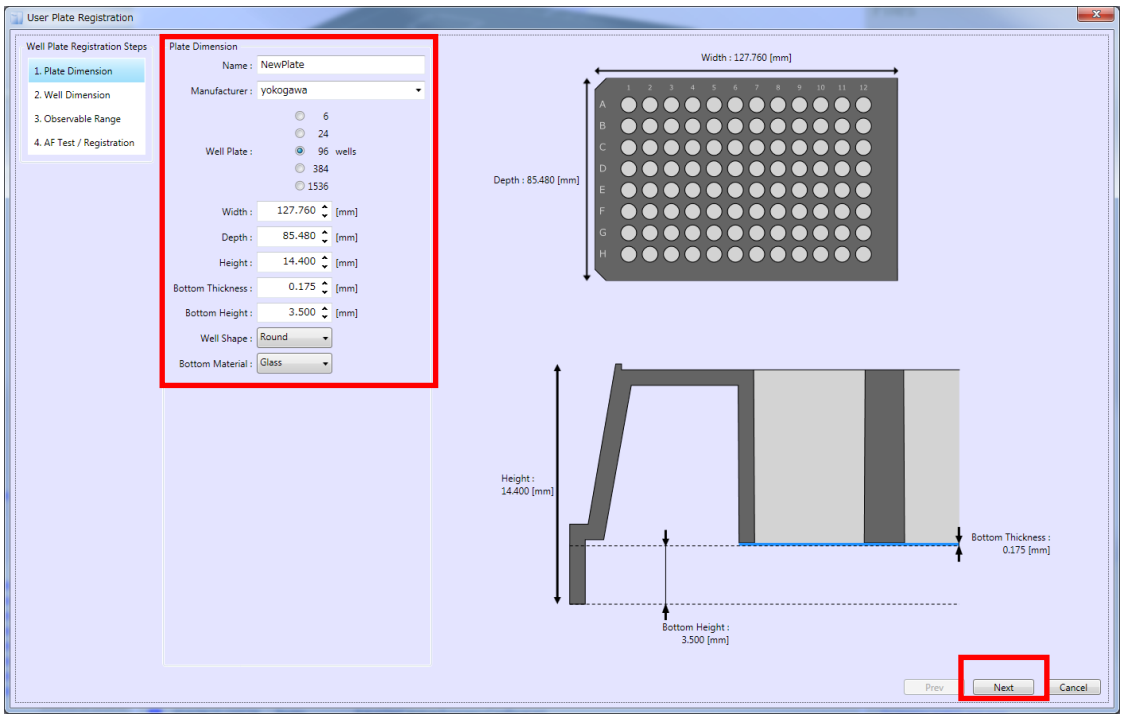
●”Register User Well Plate” cannot be selected when “Measurement Setting” window is open.
Close ”Measurement Setting” window and select “Register User Well Plate”



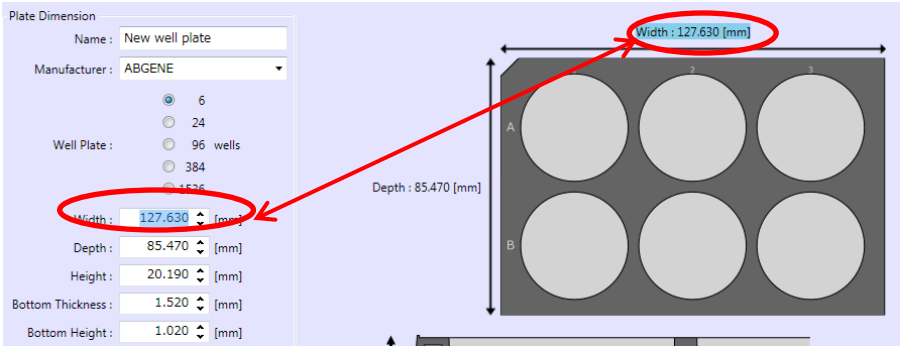
MEMO

●If water immersion objective lens is mounted, perform water supply for lens by referring page 6-43 before performing this procedure.

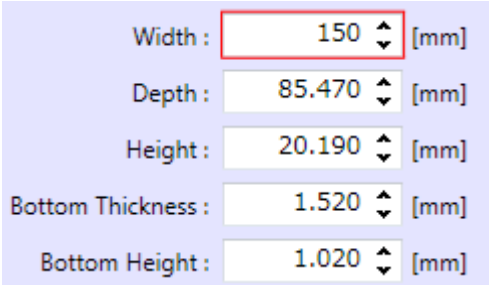
2) Input name, manufacturer, number of well, outer dimension, well shape and bottom material of well plate, then click “Next”.



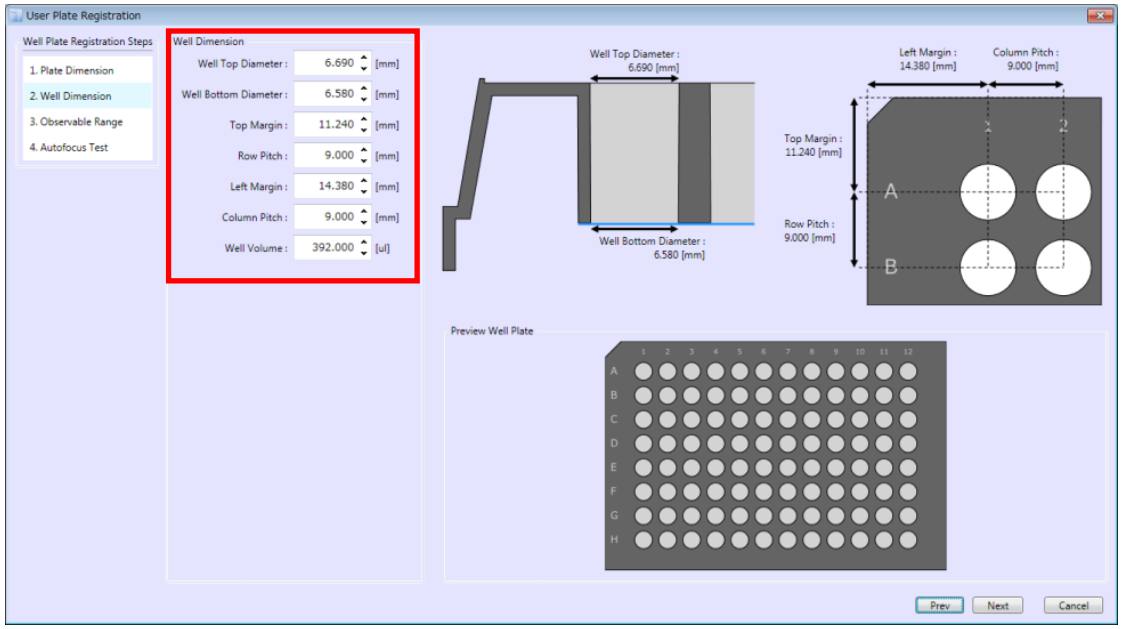
Correlated part is highlighted when dimension is input.



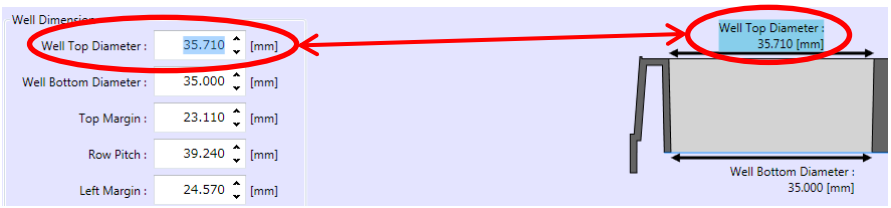
If incorrect value is input, red frame is shown and it cannot to go to next step.



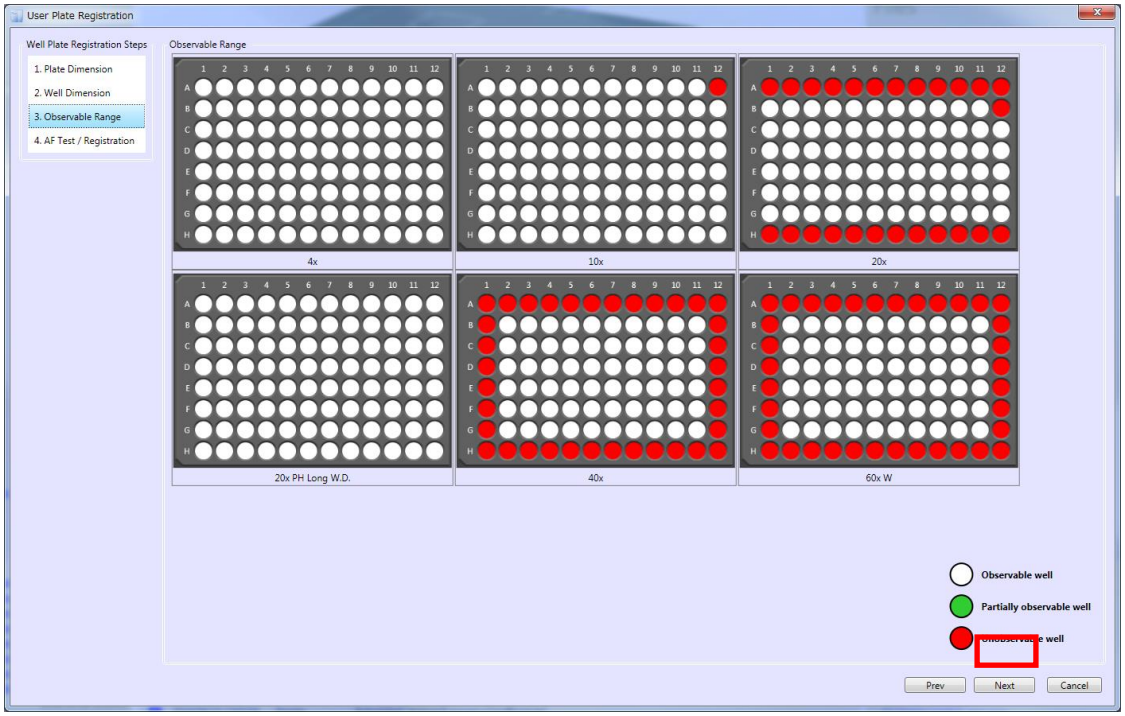
3) Input dimensions and volume of well then click “Next”.



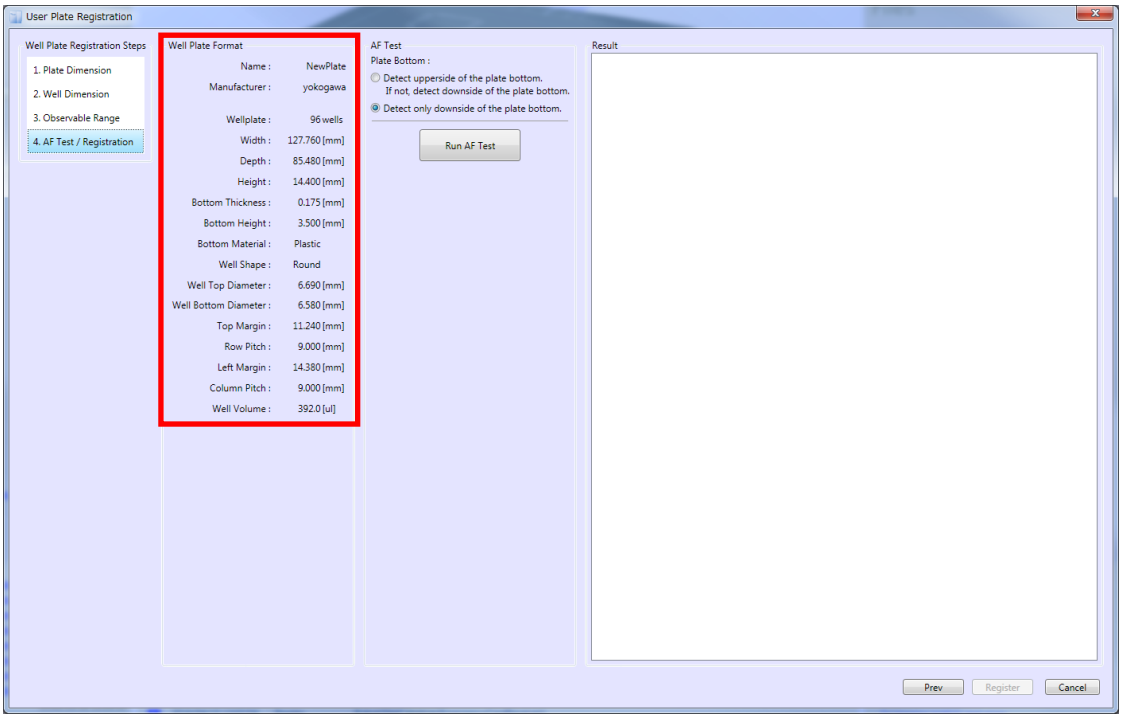
Correlated part is highlighted when dimension is input.



4) Observable range of using each objective lens is shown then click “Next”.

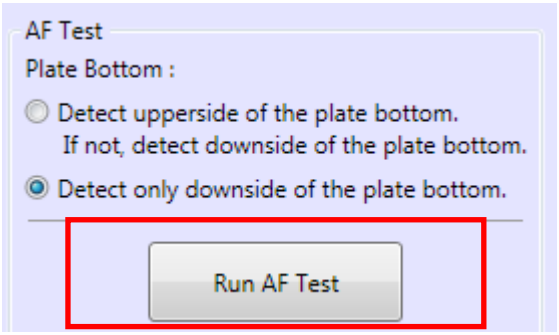


5) Confirm input well plate information.

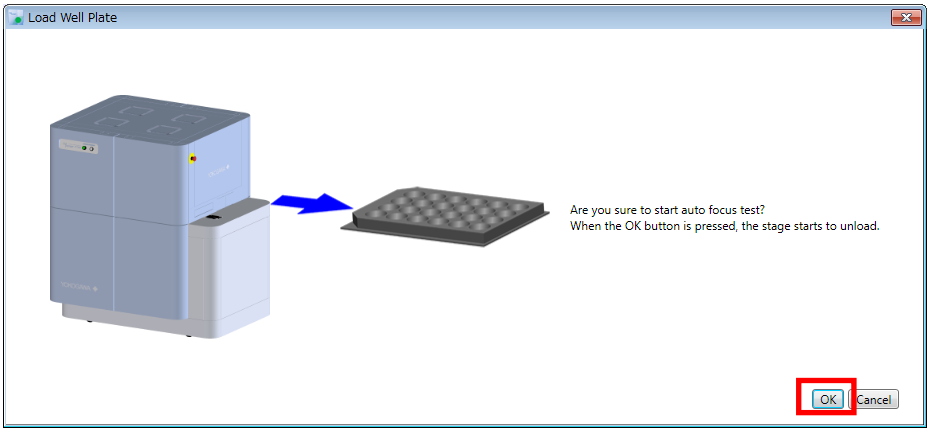


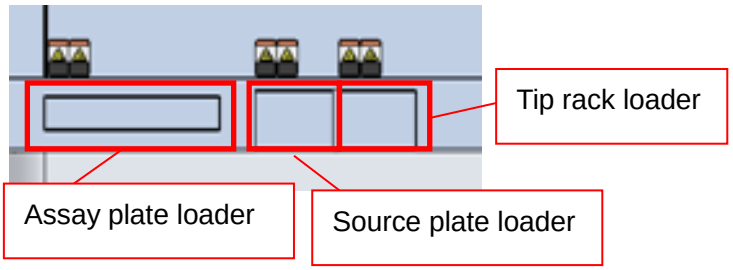
6) Test whether autofocus is available or not by using registered well plate. Normally, check “Detect upperside of the plate bottom. If not, detect down side of the plate bottom”. In following case, check “Detect only downside of the plate bottom”.
- Cell culture surface is treated specially.

Click “Run AF Test”.



7) Click “OK” to move stage to the loader exit area.

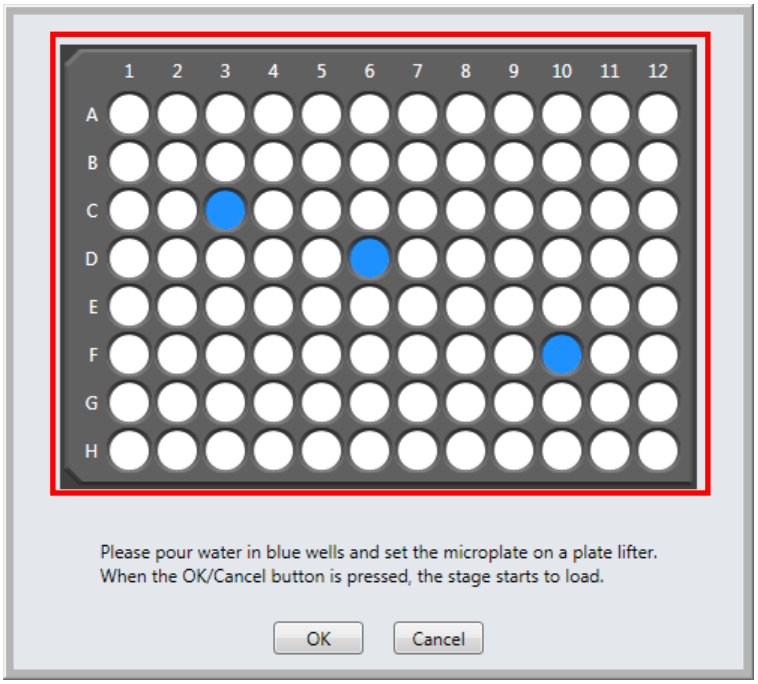


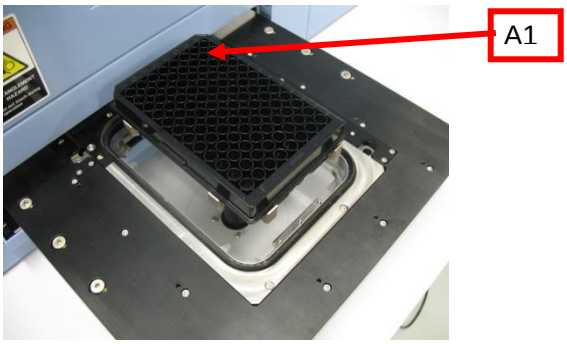


⚠ WARNING

●DO NOT touch loader exit area while moving the loaders.

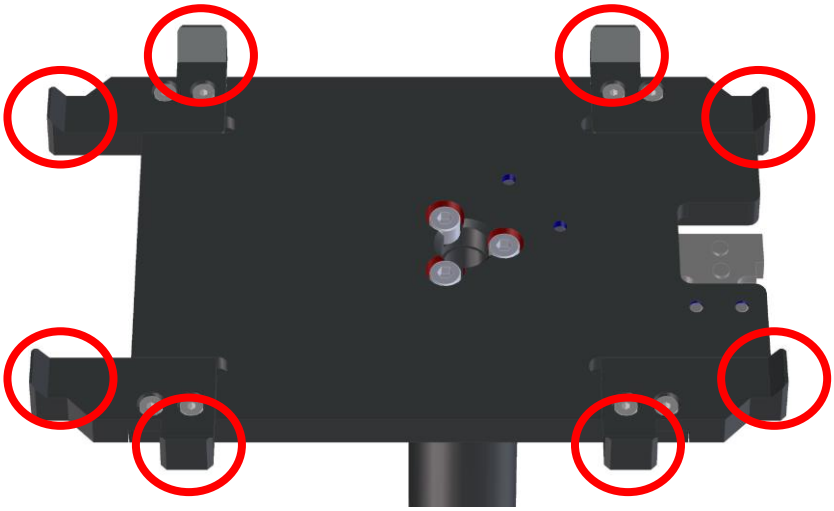
8) Prepare clean well plate, pour distillated water in designated well. Set the plate so that the well “A1” on the well plate comes to the top left-hand corner of the stage.



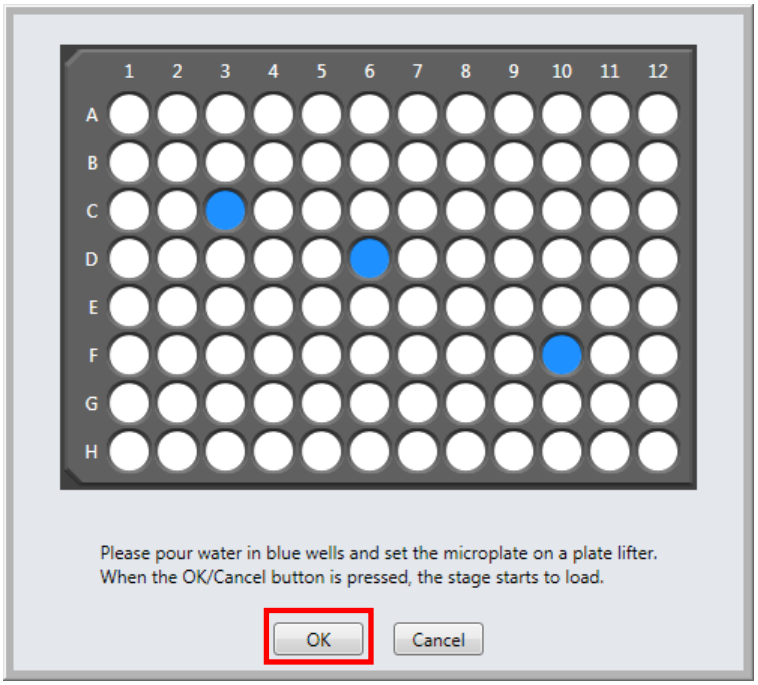


⚠ WARNING

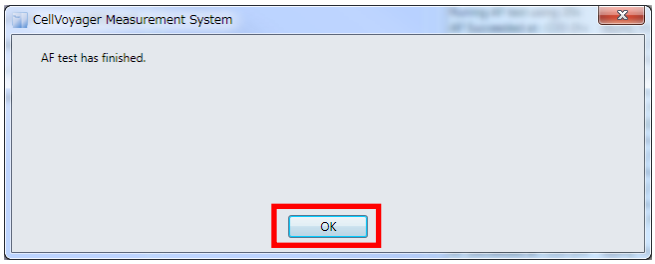
●Set the assay plate so that its eight positions align with the eight positions of the stage. Failure to do so may cause measurement unit damage.



9) Click “OK” to start AF test.



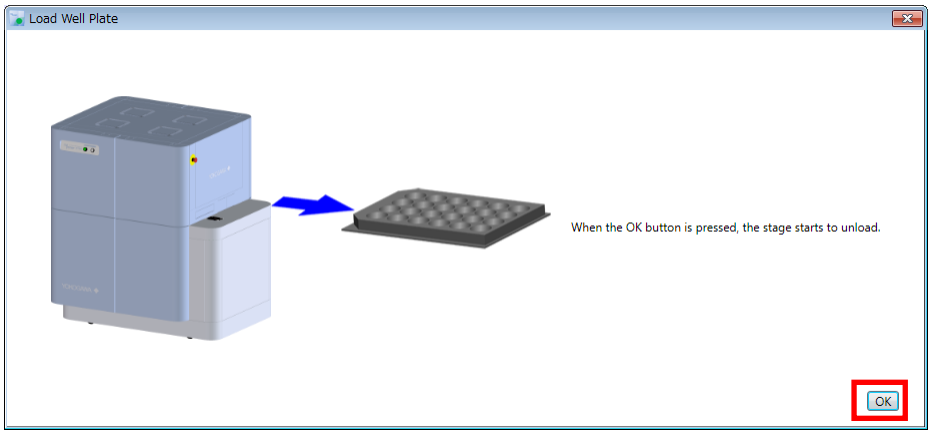
10) After AF test finishes, following window is shown. Click “OK”.



MEMO

●It becomes error if AF test fails in every time or there is gap between measurement value and user input value of “Bottom height” or “Bottom thickness”. In this case, the plate cannot be registered.
In this case, confirm that plate is clear and dimensions of plate are correct.

11) Click “OK” to move stage to the loader exit area.

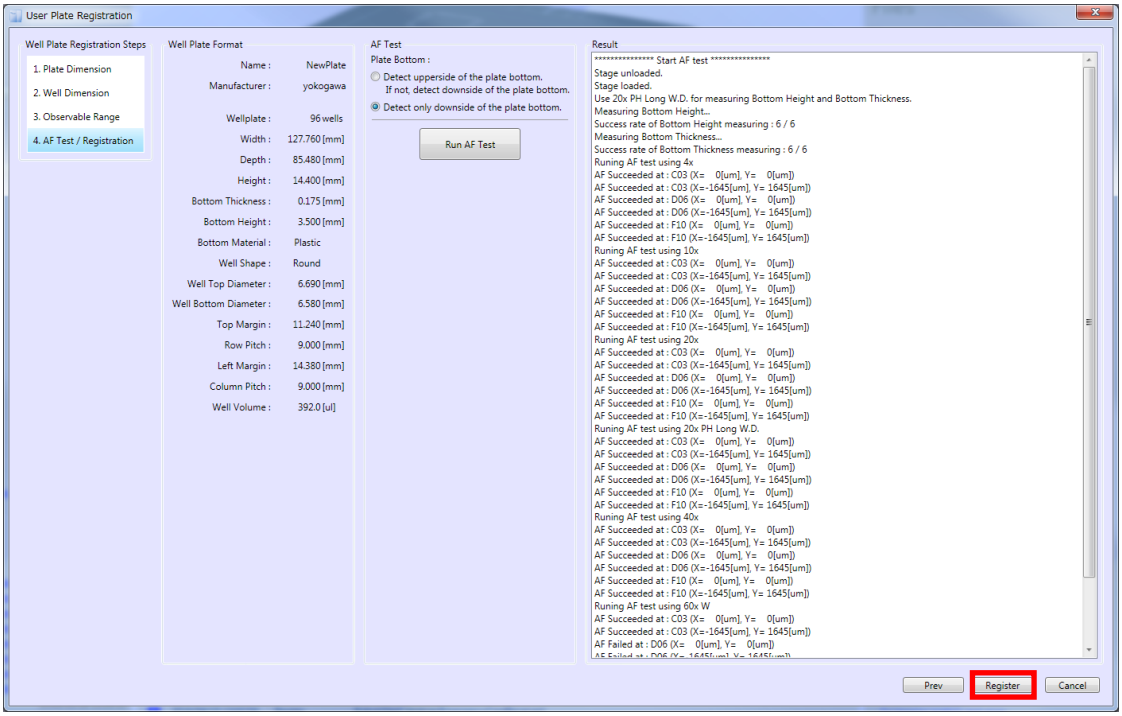


⚠ WARNING

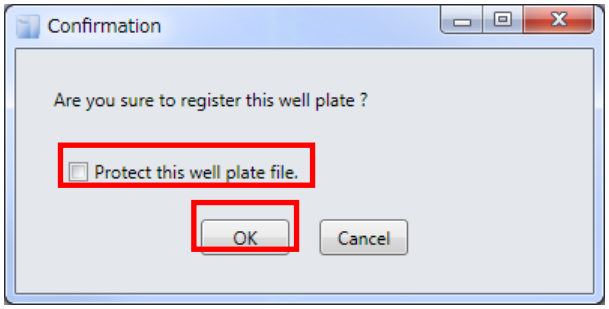
●Set the assay plate so that its eight positions align with the eight positions of the stage. Failure to do so may cause measurement unit damage.

12) If water immersion objective lens is mounted, bottom of well plate may be getting wet with water. If so, please wipe it.
Click “OK” to house stage into instrument.

13) Click “Register”.

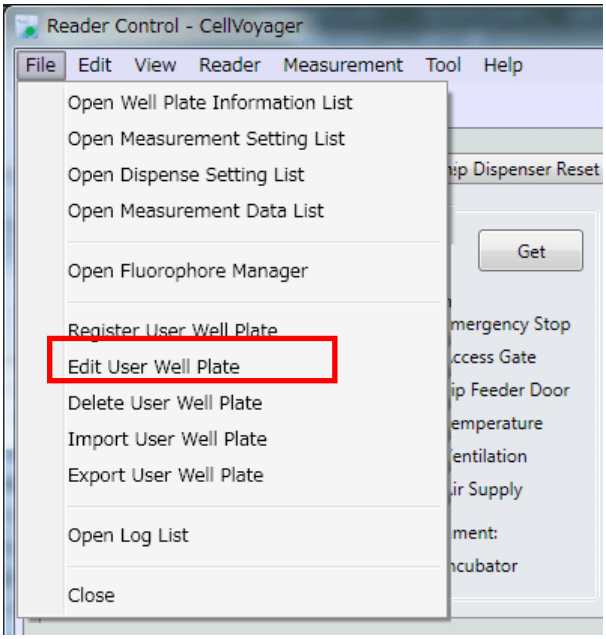


14) Confirmation window for well plate registration is shown. If you should forbid editing or deleting information file of the registered well plate, check “Protect this well plate file”. Click “OK” to finish registering.



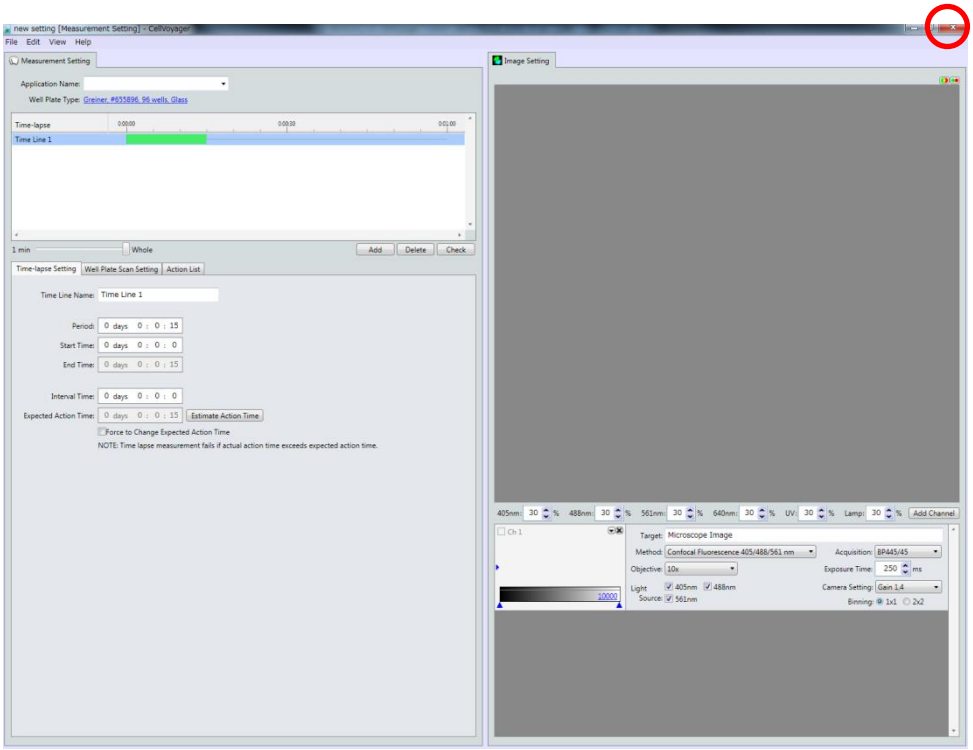
Edit User Well Plate

1) Select “File” -> “Edit User Well Plate” from menu of “Reader Control” window.



MEMO

●”Edit User Well Plate” cannot be selected when “Measurement Setting” window is open.
Close ”Measurement Setting” window and select “Edit User Well Plate”



MEMO

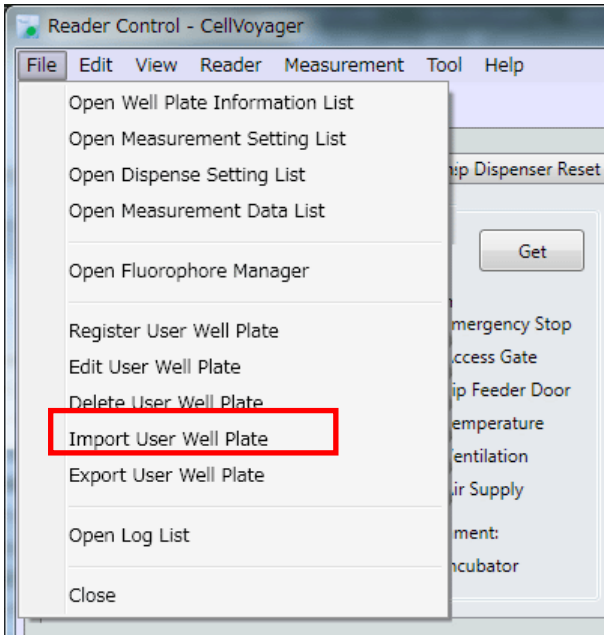
●If water immersion objective lens is mounted, perform water supply for lens by referring page 6-43 before performing this procedure.

2) Select well plate information to edit. Edit and register well plate information by referring procedure from page 5-101 to page 5-108.

- Name, Manufacturer and Well number cannot be changed in this procedure. If protected well plate information is edited, the well plate information is registered as another plate that Name, Manufacturer and Well number are same.

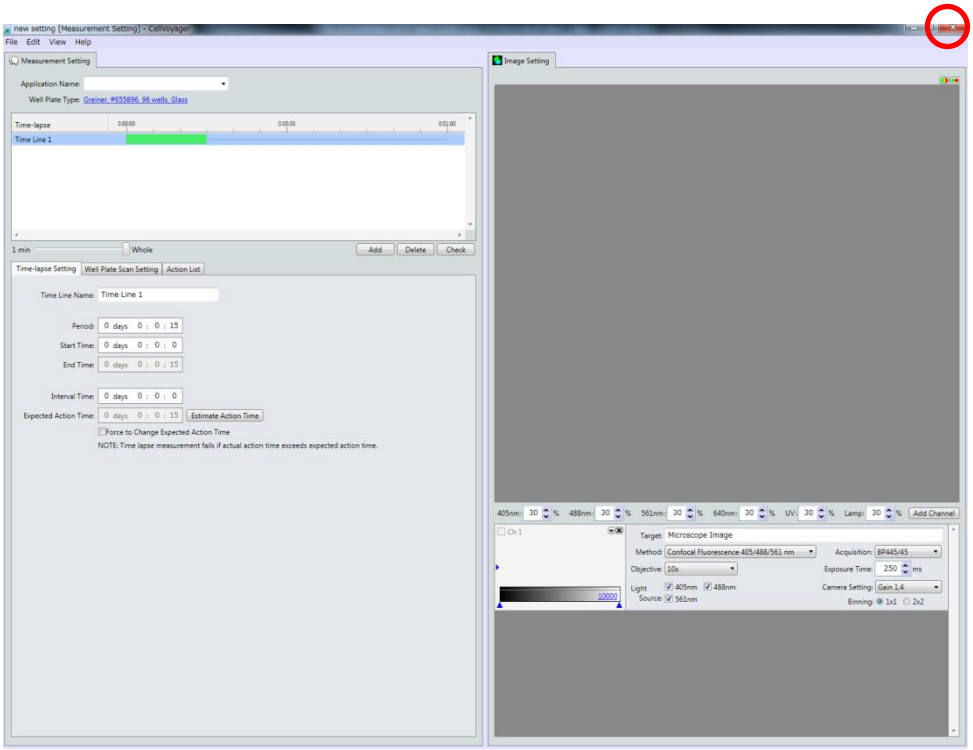
Import User Well Plate

1) Select “File” -> “Import User Well Plate” from menu of “Reader Control” window.



MEMO

●”Import User Well Plate” cannot be selected when “Measurement Setting” window is open.
Close ”Measurement Setting” window and select “Import User Well Plate”



MEMO

●If water immersion objective lens is mounted, perform water supply for lens by referring page 6-43 before performing this procedure.

2) File selecting dialog is shown. Select to load well plate information file (.wpp). Register well plate information by referring procedure from page 5-101 to page 5-108.

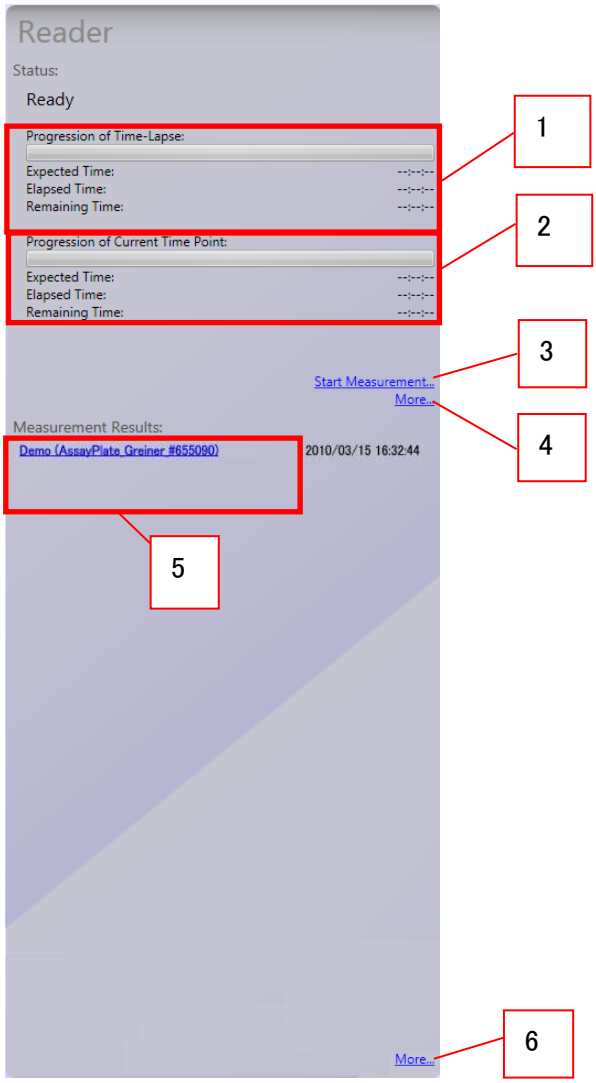
MEMO

● Name, Manufacturer and Well number cannot be changed in this procedure.

6. Explanation of Measurement Software Screens

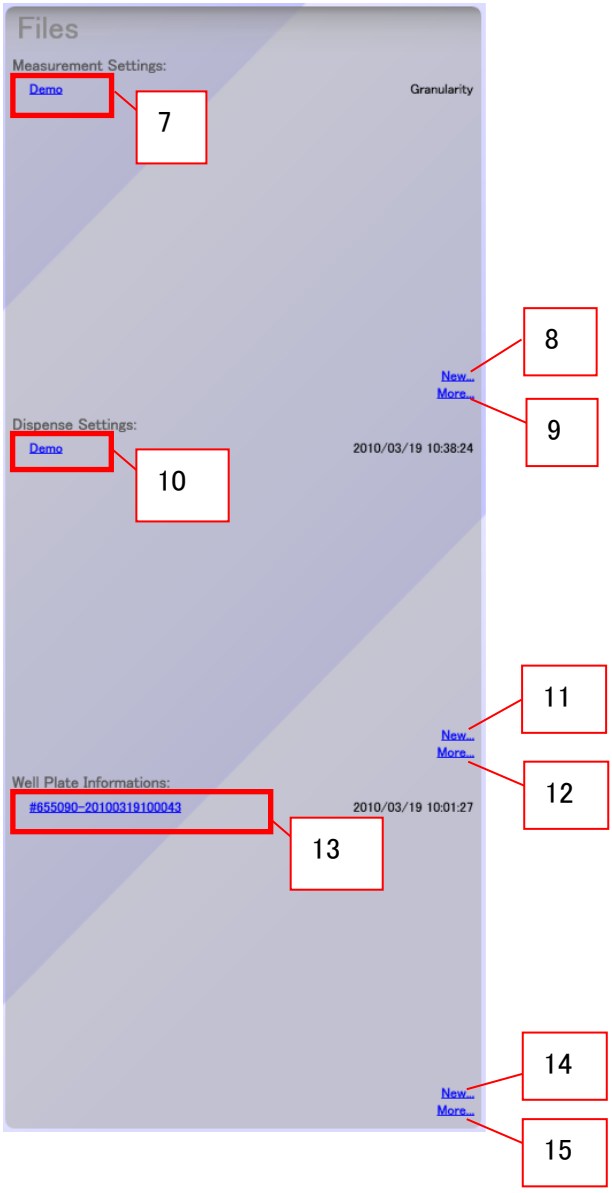
6.1. Main Screen

Reader area



1) Progress status display of time-lapse measurement
   Expected Time：Time at which all measurement will end
   Elapsed Time：Elapsed time after start of measurement
   Remaining Time：Remaining time to end of measurement
2) Progress status display of processing at one time point
   Expected Time：Time at which measurement at the time point will end
   Elapsed Time：Elapsed time after start of measurement at the time point
   Remaining Time：Remaining time to end of measurement at the time point
3) Open the screen to start measurement.
4) Open the Reader Control screen.
5) Recently measured or referenced data (Clicking here displays measured data.)
6) Browse measured data.

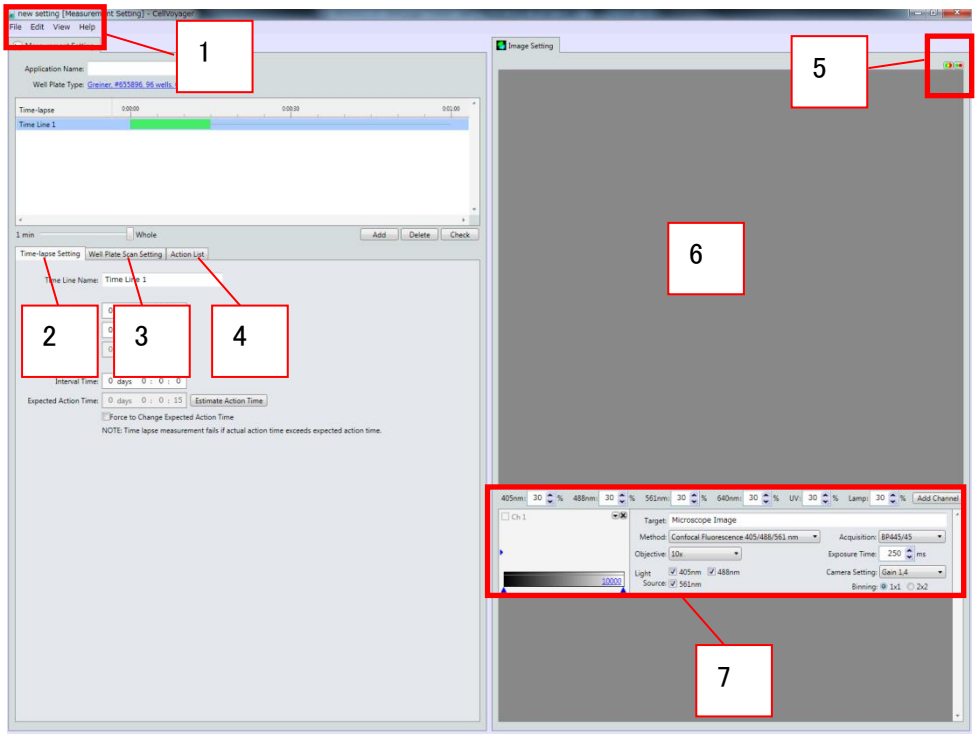
Files area



7) Recently accessed measurement settings (Clicking here opens the edit screen.)
8) Create new measurement settings.
9) Edit measurement settings.
10) Recently accessed dispensing settings (Clicking here opens the edit screen.)
11) Create new dispensing settings.
12) Edit dispensing settings.
13) Recently accessed well plate information (Clicking here opens the edit screen.)
14) Create new well plate information.
15) Edit well plate information.

6.2. Measurement Setting File Screen

Main Measurement Setting File Screen



1) Menus

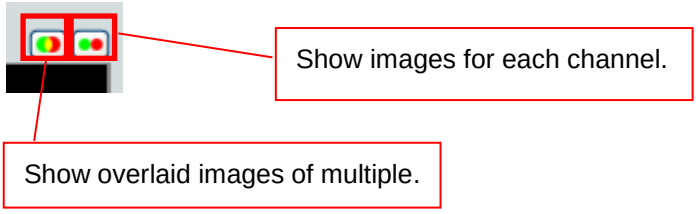
File menu	Explanation
Save	Save the measurement setting file under the same name.
Save As	Save the measurement setting file under a new name.
Close	Close the measurement setting file.

Edit menu	Explanation
Undo	Undo the last operation.
Redo	Redo the last operation.
Cut	Cut the selected item.
Copy	Copy the selected item.
Paste	Paste the selected item.

View menu	Explanation
Time Line Setting	Open the Time-lapse Setting tab.
Plate Scan Setting	Open the Well Plate Scan Setting tab.
Action List Setting	Open the Action List tab.
Show Overlay Images	Open the preview screen showing overlaid images of multiple channels.
Show Tile Images	Open the preview screen showing images for each channel.

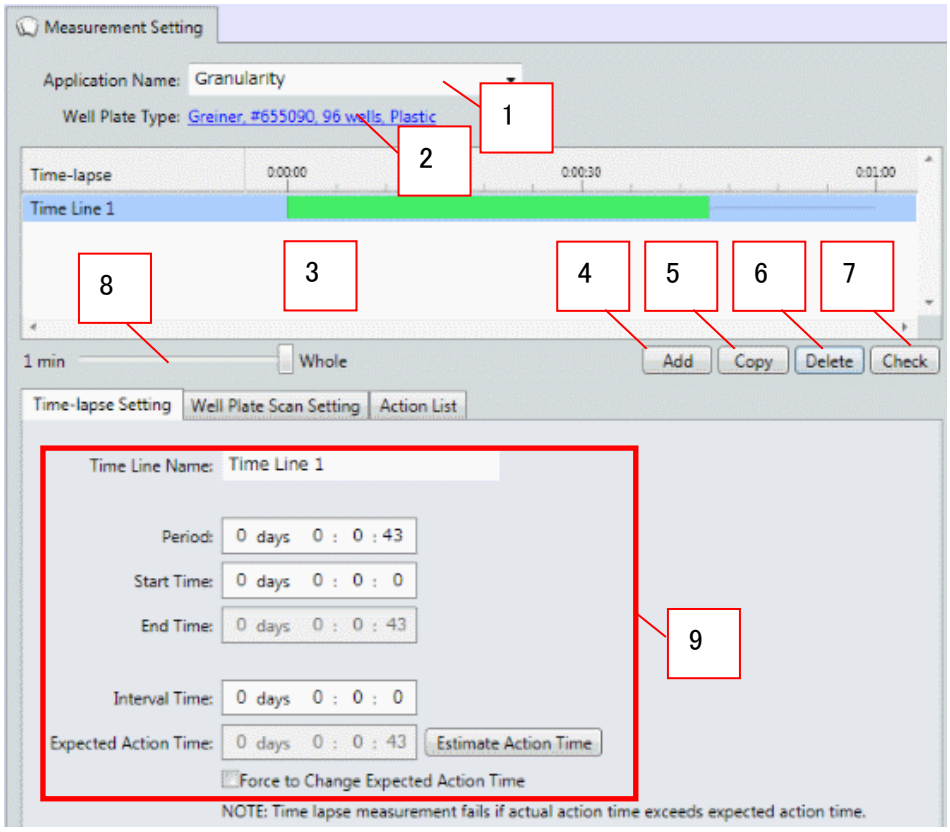
Help menu	Explanation
About	Show the version information of the measurement software.

2) Time-lapse Setting tab
3) Well Plate Scan Setting tab
4) Action List tab
5) Select the display format for channel images.



6) Preview screen
7) Imaging channel setting screen

Time-lapse Setting Screen

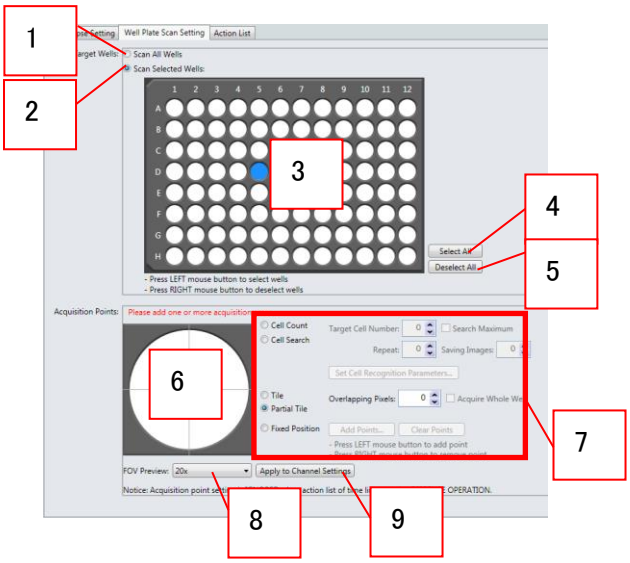


1) Application name
2) Well plate product name
   The well plate product can be changed by clicking here.
3) Time-line display area
4) Add a time line.
5) Copy the selected time line and add to the last of time line list.
6) Delete the selected time line.
7) Automatically and optimally arrange the time lines overlapping along the time axis
8) Change the display scale for time line.

9) Time-line setting items

Item	Explanation
Time Line Name	Specify the name of the time line.
Period	Specify the period of the time line, or duration after the start time until the time line ends.
Start Time	Specify the start time of the time line as, or the duration after the start of measurement until the first time point starts.
End Time	The end time of the time line is shown.
Interval Time	Set the duration after a time point ends until the next time point starts.
Expected Action Time	The expected processing time defined on the Action List tab is shown.
Estimate Action Time	The predicted processing time defined on the Action List tab is calculated when this button is clicked.
Force to Change Expected Action Time	The Expected Action Time field can be entered.

Well Plate Scan Setting Screen



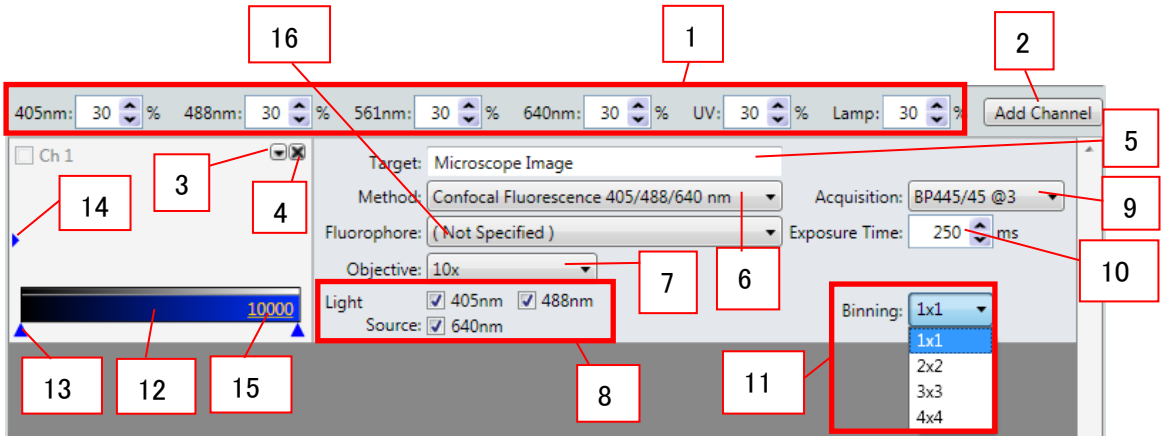
1) Measure all the wells.
2) Select wells to be measured.
3) Display area of wells to be measured
4) Select all the wells.
5) Unselect all the wells.
6) Imaging view field display area
7) Set the imaging view field.

Item	Explanation
Cell Count	An image is captured repeatedly while moving through the acquisition points, until the cell count entered in “Cell Number” is reached. Once the number of repetitions reaches the value in “Repeat,” the system moves to the next well.
Cell Search	Move to the next acquisition point repeatedly until the value in “Repeat” is reached, to find the acquisition point associated with the largest cell count.
Target Cell Number	Specify the cell count.
Repeat	Specify the number of images to be captured for the same well.
Search Maximum	Save the images one by one, starting from the image associated with the largest cell count.
Saving Images	Specify the number of images to be saved.
Set Cell Recognition Parameters	Open the screen for setting the cell recognition algorithm.
Tile	Images are captured in a tiled manner. (whole region of well)
Partial Tile	Images are captured in a tiled manner. (desired region(s) of well)
Overlapping Pixels	Specify the number of overlapping pixels between images for tiled imaging.

Acquire Whole Well	Whole-well imaging is performed.
Fixed Position	Directly specify the positions of acquisition points.
Add Points	Specify the number and pitch of acquisition points, etc.
Clear Points	Clear the acquisition points.

8) Show the view of specified lens magnification.
9) Apply the lens magnification specified with “FOV Preview” to “Imaging Channel”. (Refer to 5.5)

Imaging Channel Setting Screen

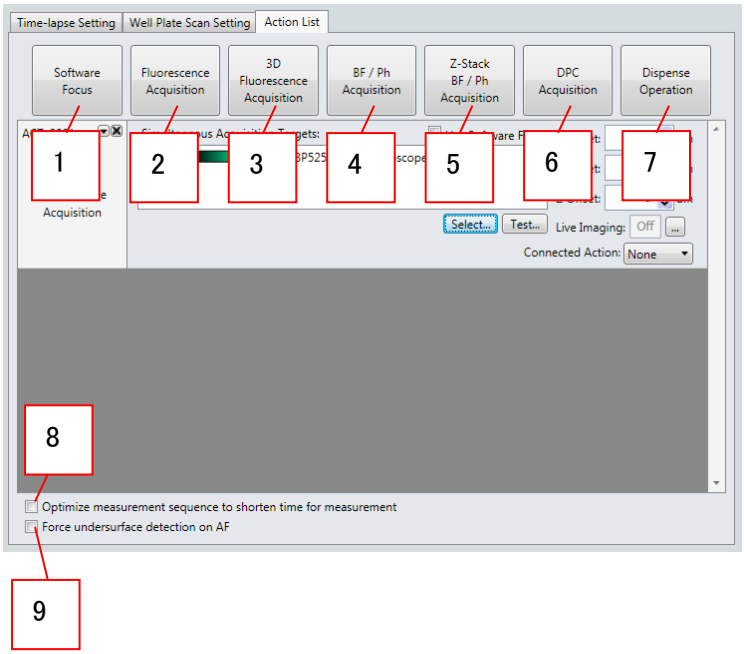


1) Set the laser power.
2) Create a new imaging channel.
3) Move the channel list.

Item	Explanation
Cut This Channel	Cut a channel list.
Copy This Channel	Copy a channel list.
Paste Channel Above	Paste the selected channel list above the one currently selected.
Paste Channel Below	Paste the selected channel list below the one currently selected.

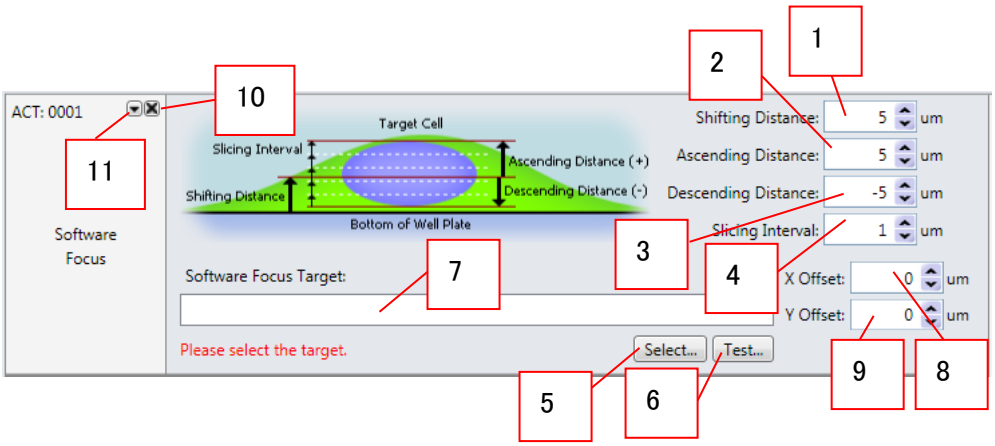
4) Delete the imaging channel.
5) Imaging target name
6) Select the optical system used for imaging.
7) Select the magnification factor.
8) Select the laser.
9) Select the fluorescence filter.
10) Camera exposure time
11) Camera binning
12) Adjust the image color.
13) Contrast bar
14) Expand the screen for contrast adjustment.
15) Select the maximum intensity for contrast adjustment.
16) Select fluorophore.

Action List Screen



1) Add the Action for Software Focus.
2) Add the Action for Fluorescence Acquisition.
3) Add the Action for 3D Fluorescence Acquisition.
4) Add the Action for Bright-field/Phase-contrast Acquisition.
5) Add the Action for Z-Stack Bright-field/Phase-contrast Acquisition.
6) Add the Action for DPC Acquisition.
7) Add the Action for Dispense Operation.
8) Optimize measurement sequence. (Refer to 7.3)
   Checked : Optical switching is optimized to acquire images with the minimum time.
   Unckecked : Optical switching is performed to acquire images at same time point.
9) Set to force undersurface detection on AF.
   (Set when performing autofocus during high-speed time-lapse imaging)

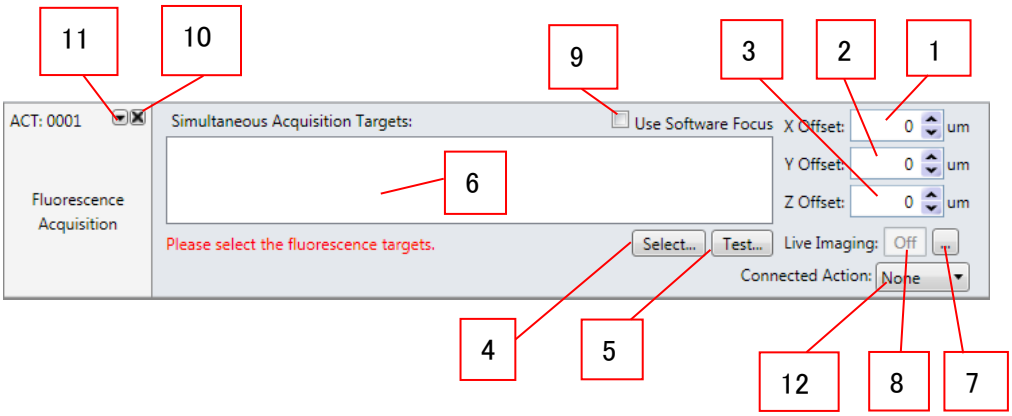
Software Focus Screen



1) Reference plane of software focus
2) Upper limit of software focus
3) Lower limit of software focus
4) Z-axis pitch of software focus
5) Select the target channel for software focus.
6) Open the test preview screen.
7) The target channel for software focus is shown.
8) Fine-tune the X-axis of image positions.
9) Fine-tune the Y-axis of image positions.
10) Close the software focus screen.
11) Move the action list.

Item	Explanation
Cut This Action	Cut an action
Copy This Action	Copy an action
Paste Action Above	Paste the selected action above the one currently selected.
Paste Action Below	Paste the selected action below the one currently selected.

Fluorescence Acquisition Screen

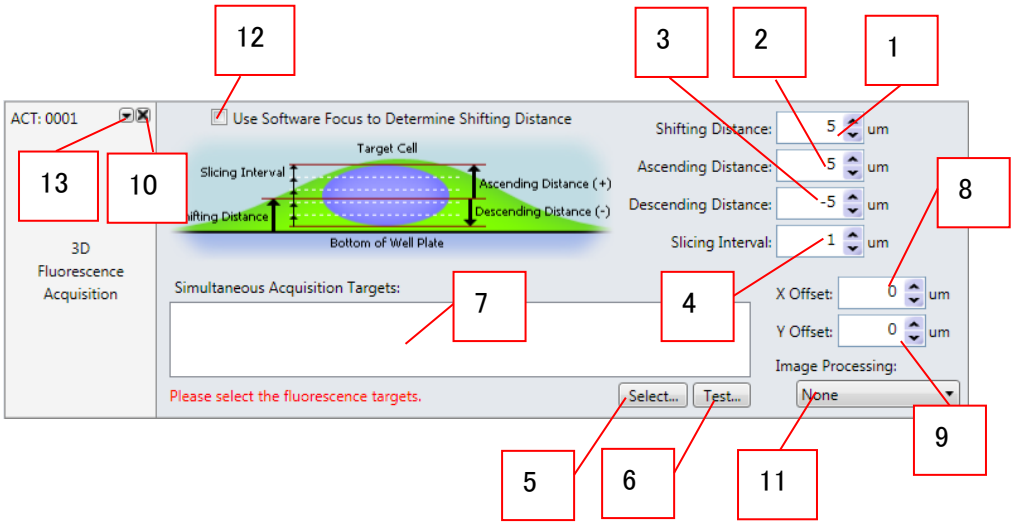


1) Fine-tune the X-axis of image positions.
2) Fine-tune the Y-axis of image positions.
3) Fine-tune the Z-axis of image positions.
4) Select the target channel for fluorescence imaging.
5) Open the test preview screen.
6) Show the target channel for fluorescence imaging.
7) Open the screen for setting high-speed time-lapse imaging.
8) Use condition of high-speed time-lapse imaging - On：Use, Off：Do not use
9) Perform fluorescence imaging by using software focus plane as a reference.
10) Close the fluorescence imaging screen.
11) Move the action list.

Item	Explanation
Cut This Action	Cut an action
Copy This Action	Copy an action
Paste Action Above	Paste the selected action above the one currently selected.
Paste Action Below	Paste the selected action below the one currently selected.

12) Set the connection to high-speed time-lapse imaging (refer to 7.15).

3D Fluorescence Acquisition Screen



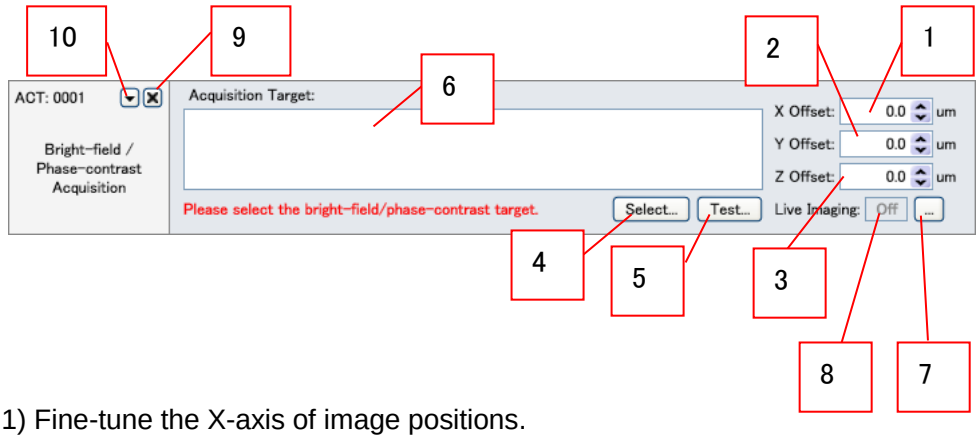
1) Reference plane of 3D imaging
2) Upper limit of 3D imaging
3) Lower limit of 3D imaging
4) Z-axis pitch of 3D imaging
5) Select the target channel for 3D imaging.
6) Open the test preview screen.
7) The target channel for 3D imaging is shown.
8) Fine-tune the X-axis of image positions.
9) Fine-tune the Y-axis of image positions.
10) Close the 3D imaging screen.
11) Select an output method for Z-stack images.

Item	Explanation
None	Acquired all Z images are saved.
Maximum	MIP images by Z-axis direction are saved.
Minimum	MinIP images by Z-axis direction are saved.
Average	AIP images by Z-axis direction are saved.
Sum	SUM images by Z-axis direction are saved.

12) Perform 3D imaging by using the software focus plane as a reference.
13) Move the action list.

Item	Explanation
Cut This Action	Cut an action
Copy This Action	Copy an action
Paste Action Above	Paste the selected action above the one currently selected.
Paste Action Below	Paste the selected action below the one currently selected.

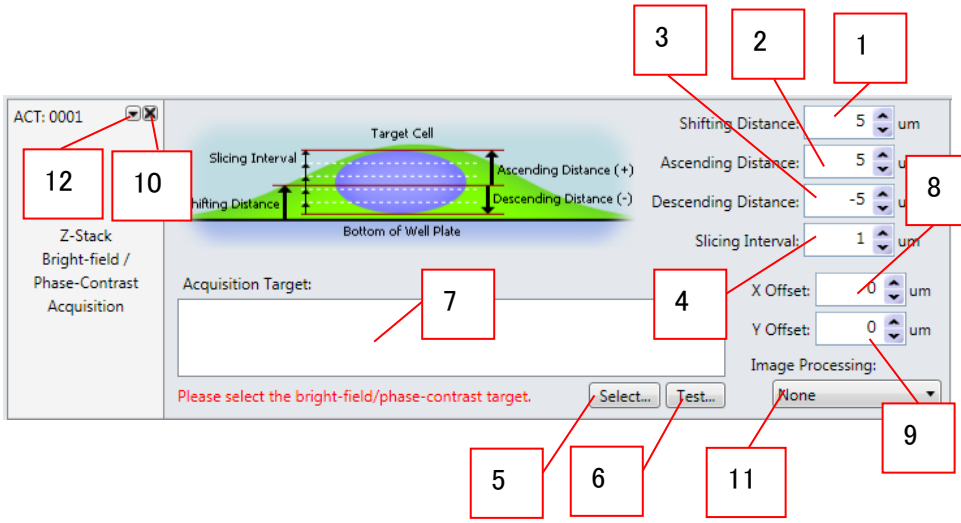
Bright Field/Phase Contrast Acquisition Screen



1) Fine-tune the X-axis of image positions.
2) Fine-tune the Y-axis of image positions.
3) Fine-tune the Z-axis of image positions.
4) Select the target channel for bright field/phase contrast imaging.
5) Open the test preview screen.
6) The target channel for bright field/phase contrast imaging is shown.
7) Open the screen for setting high-speed time-lapse imaging.
8) Use condition of high-speed time-lapse imaging - On：Use, Off：Do not use
9) Close the Bright field/Phase Contrast Acquisition screen.
10) Move the action list.

Item	Explanation
Cut This Action	Cut an action
Copy This Action	Copy an action
Paste Action Above	Paste the selected action above the one currently selected.
Paste Action Below	Paste the selected action below the one currently selected.

Z-Stack Bright-field/Phase-contrast Acquisition



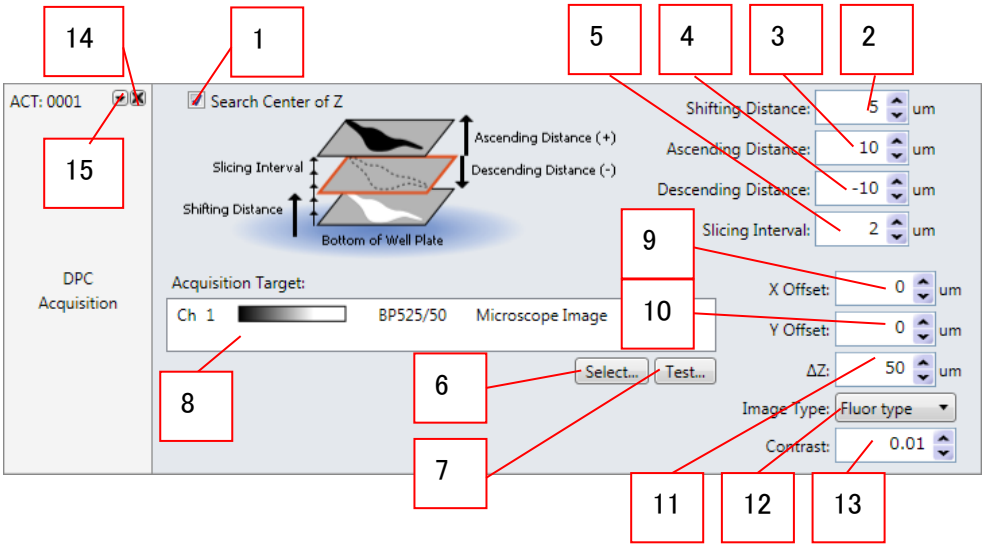
1) Reference plane of Z-stack imaging
2) Upper limit of Z-stack imaging
3) Lower limit of Z-stack imaging
4) Z-axis pitch of Z-stack imaging
5) Select the target channel for Z-stack imaging.
6) Open the test preview screen.
7) The target channel for Z-stack imaging is shown.
8) Fine-tune the X-axis of image positions.
9) Fine-tune the Y-axis of image positions.
10) Close the Z-Stack Bright-field/Phase-contrast Acquisition screen.
11) Select an output method for Z-stack images.

Item	Explanation
None	Acquired all Z images are saved.
Maximum	MIP images by Z-axis direction are saved.
Minimum	MinIP images by Z-axis direction are saved.
Average	AIP images by Z-axis direction are saved.
Sum	SUM images by Z-axis direction are saved.

12) Move the action list.

Item	Explanation
Cut This Action	Cut an action
Copy This Action	Copy an action
Paste Action Above	Paste the selected action above the one currently selected.
Paste Action Below	Paste the selected action below the one currently selected.

DPC Acquisition



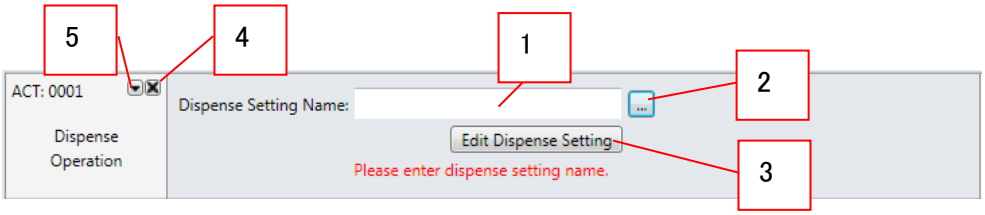
1) Select performing automatic DPC reference position search
2) Reference plane of automatic DPC reference position search
※In case that “Search Center of Z” is unchecked, this means DPC reference position
3) Upper limit of automatic DPC reference position search
4) Lower limit of automatic DPC reference position search
5) Z-axis pitch of automatic DPC reference position search
6) Select the target channel for DPC imaging.
7) Open the test preview screen.
8) The target channel for DPC imaging is shown.
9) Fine-tune the X-axis of image positions.
10) Fine-tune the Y-axis of image positions.
11) Set ΔZ (Z distance of multiple bright field images which are origins of DPC image)
12) Select the type of DPC image

Item	Explanation
Fluor type	Fluorescence like DPC image.
Phase type	Phase contrast like DPC image.

13) Set contrast of DPC image to output.
14) Delete action of DPC Acquisition.
15) Move the action list.

Item	Explanation
Cut This Action	Cut an action
Copy This Action	Copy an action
Paste Action Above	Paste the selected action above the one currently selected.
Paste Action Below	Paste the selected action below the one currently selected.

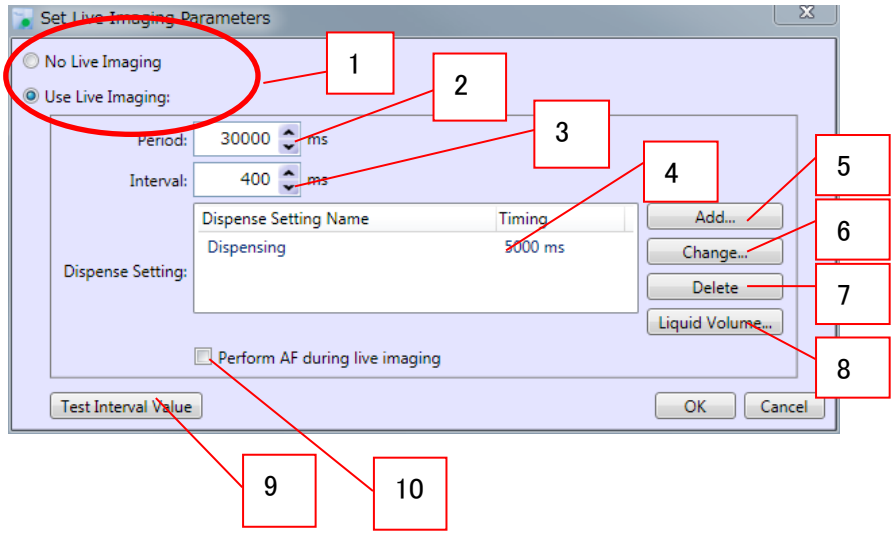
Dispense Operation Screen



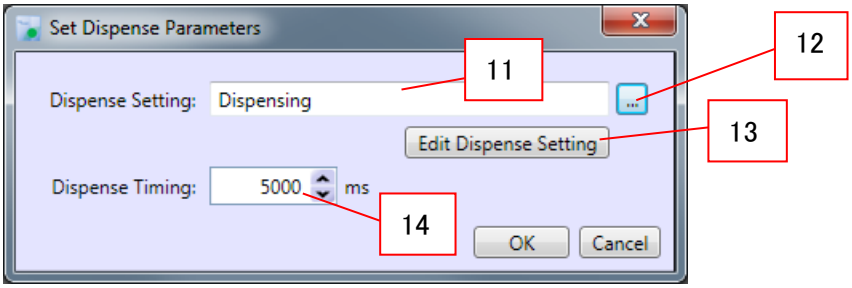
1) The dispensing setting file is shown.
2) Select an applicable dispensing setting file.
3) Edit the dispensing setting file to have been selected.
4) Close the dispensing screen.
5) Move the action list.

Item	Explanation
Cut This Action	Cut an action
Copy This Action	Copy an action
Paste Action Above	Paste the selected action above the one currently selected.
Paste Action Below	Paste the selected action below the one currently selected.

High-speed Time-lapse Setting Screen

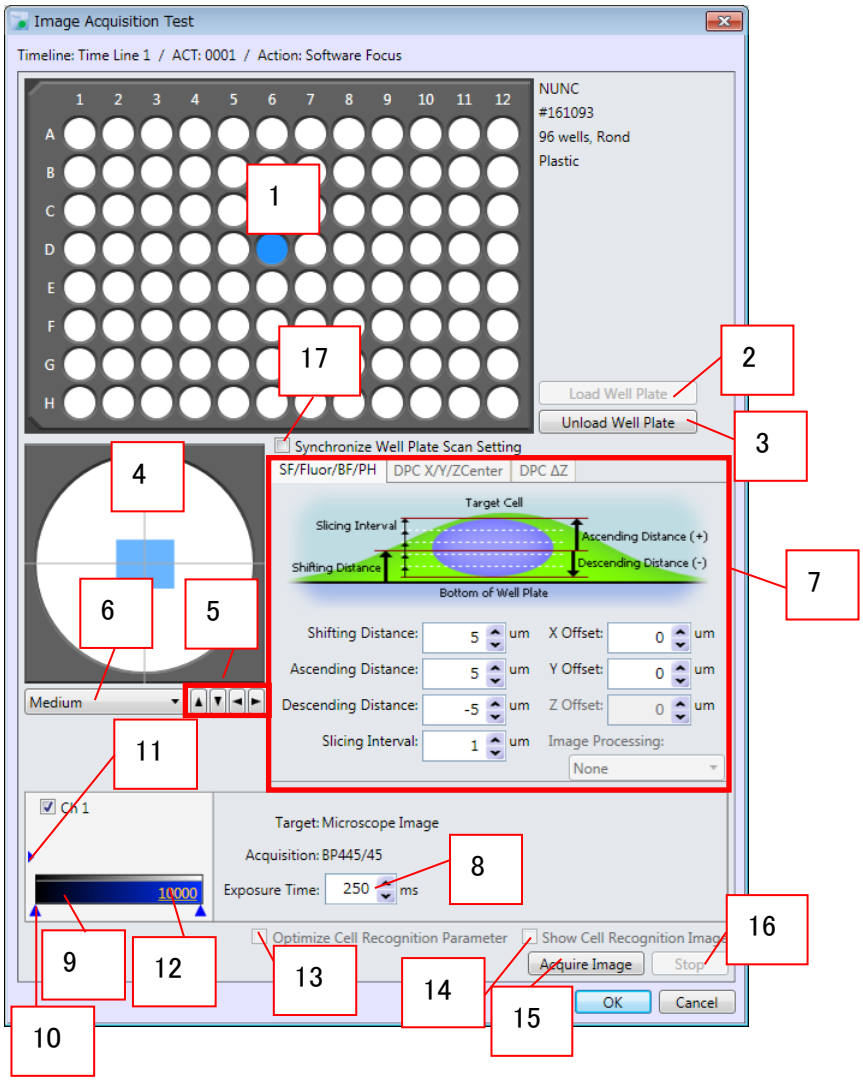


1) Selection of whether to use (Use Live Imaging) or not use (No Live Imaging) high-speed time-lapse imaging.
2) Period of high-speed time-lapse imaging
3) Interval of high-speed time-lapse imaging
4) Dispensing setting for high-speed time lapse imaging
5) Specify dispensing setting file (Set this item if dispensing is performed.)
6) Change dispensing setting file to have been registered.
7) Delete dispensing setting file to have been registered.
8) Display the Liquid Volume screen. (Refer to 8.4.)
9) Test to calculate “Interval” from the setting parameters such as exposure time, etc.
10) Selection of whether to perform autofocus during high-speed time-lapse imaging.



11) Dispensing setting file
12) Select dispensing setting file.
13) Edit the dispensing setting file to have been selected.
14) Timing at which to drip reagent after the start of high-speed time-lapse imaging (Set this item if dispensing is performed.)

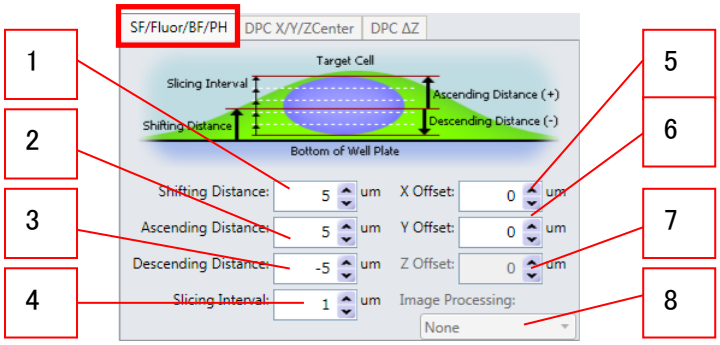
Image Acquisition Test Setting Screen



1) Select the well used in the image-acquisition test.
2) Load the well plate.
3) Remove the well plate.
4) Select the view field for image-acquisition test.
5) Move the view field for image-acquisition test.
6) Select the moving speed of the view field for image-acquisition test.
7) Set imaging conditions. (Detail is shown in next page)
8) Camera exposure time
9) Adjust the image color.
10) Contrast bar
11) Expand the screen for contrast adjustment.
12) Select the maximum intensity for contrast adjustment.
13) Optimize the set values of recognition algorithm.
14) Show binary images.
15) Start the image-acquisition test.
16) Stop the image-acquisition test.
17) Reflect the wells observed on the preview screen, and the corresponding view field, in the Acquisition Points settings.

Image Acquisition Setting Screen (Acquisition Test Setting)

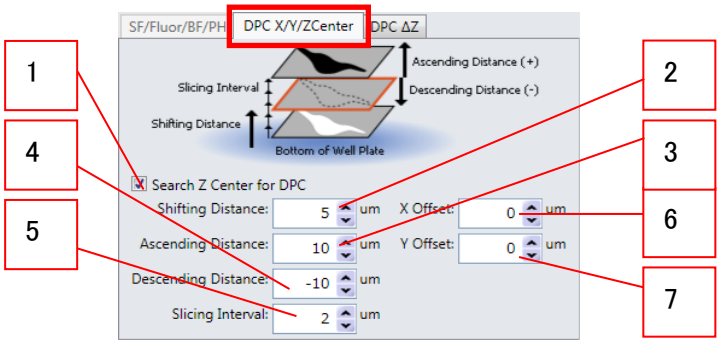
<Acquisition position setting
(Software Focus, Fluorescence/ BF/ PH Acquisition)>



1) Reference plane of Z-stack imaging
2) Upper limit of Z-stack imaging
3) Lower limit of Z-stack imaging
4) Z-axis pitch of Z-stack imaging
5) Fine-tune the X-axis of image positions.
6) Fine-tune the Y-axis of image positions.
7) Fine-tune the Z-axis of image positions.
8) Select an output method for Z-stack images.

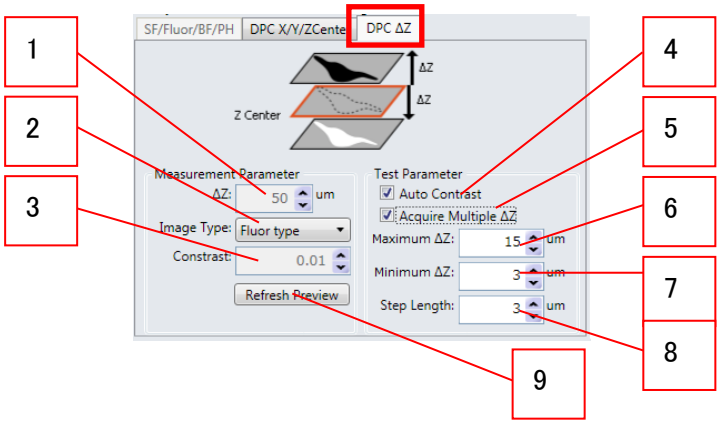
Item	Explanation
None	Acquired all Z images are saved.
Maximum	MIP images by Z-axis direction are saved.
Minimum	MinIP images by Z-axis direction are saved.
Average	AIP images by Z-axis direction are saved.
Sum	SUM images by Z-axis direction are saved.

<Acquisition position setting (DPC Acquisition)>



1) Select performing automatic DPC reference position search
2) Reference plane of automatic DPC reference position search
※In case that “Search Center of Z” is unchecked, this means DPC reference position
3) Upper limit of automatic DPC reference position search
4) Lower limit of automatic DPC reference position search
5) Z-axis pitch of automatic DPC reference position search
6) Fine-tune the X-axis of image positions.
7) Fine-tune the Y-axis of image positions.

<Image property setting (DPC Acquisition)>



1) Set ΔZ (Z distance of multiple bright field images which are origins of DPC image)
2) Select the type of DPC image

Item	Explanation
Fluor type	Fluorescence like DPC image.
Phase type	Phase contrast like DPC image.

3) Set contrast of DPC image to output.
4) Set auto contrast.
5) Select performing multiple ΔZ test acquisition.
6) Maximum ΔZ of multiple ΔZ test acquisition.
7) Minimum ΔZ of multiple ΔZ test acquisition.
8) ΔZ pitch of multiple ΔZ test acquisition.
9) Apply new contrast value to test images.

6.3. Dispensing Setting File Screen

Overview of Dispensing Setting File Screen

1) Menus

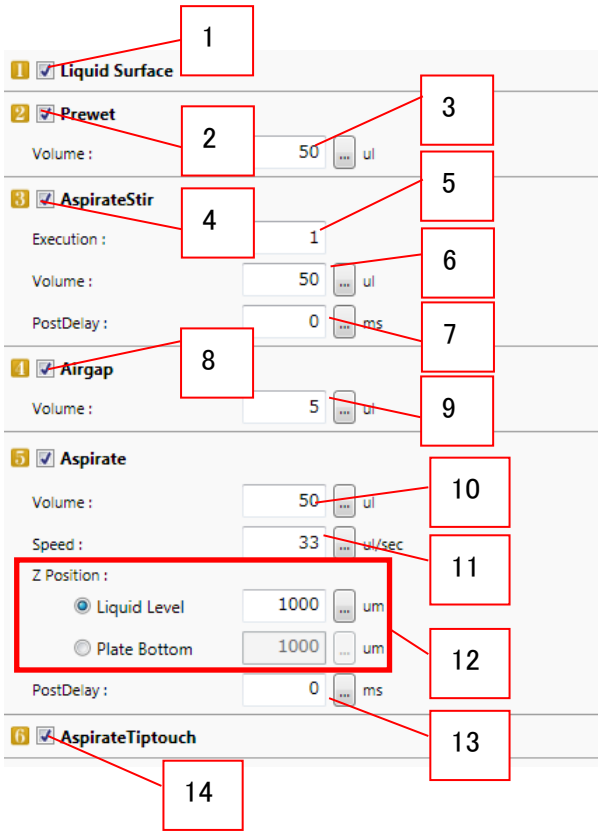
File	Explanation
Save	Save the dispensing setting file under the same name.
Save As	Save the dispensing setting file under a new name.
Close	Close the dispensing setting file.

View	Explanation
Dispense Mapping	Open the Dispense Mapping screen.
Dispense Simulation	Open the Dispense Simulation screen.
Basic Dispense Setting	Open the Basic Dispense Setting screen.
Advanced Dispense Setting	Open the Advanced Dispense Setting screen.

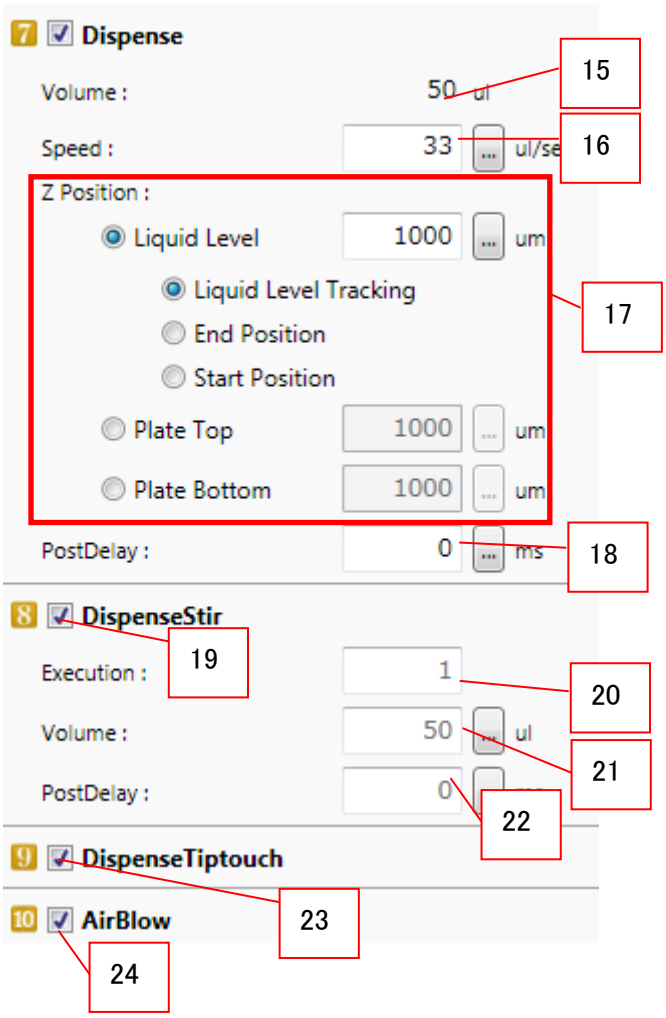
Simulation	Explanation
Start Simulation	Start simulation based on the dispensing settings.
Clear Simulation Log	Clear the simulation log.

2) Show the list of dispensing settings.
3) Dispensing settings for each well
4) Plate view screen
5) Add a well plate to the source plate view screen.
6) Add a well plate to the assay plate view screen.
7) Associate the source and assay plates.
8) Delete the association of source and assay plates.

Basic Setting Screen

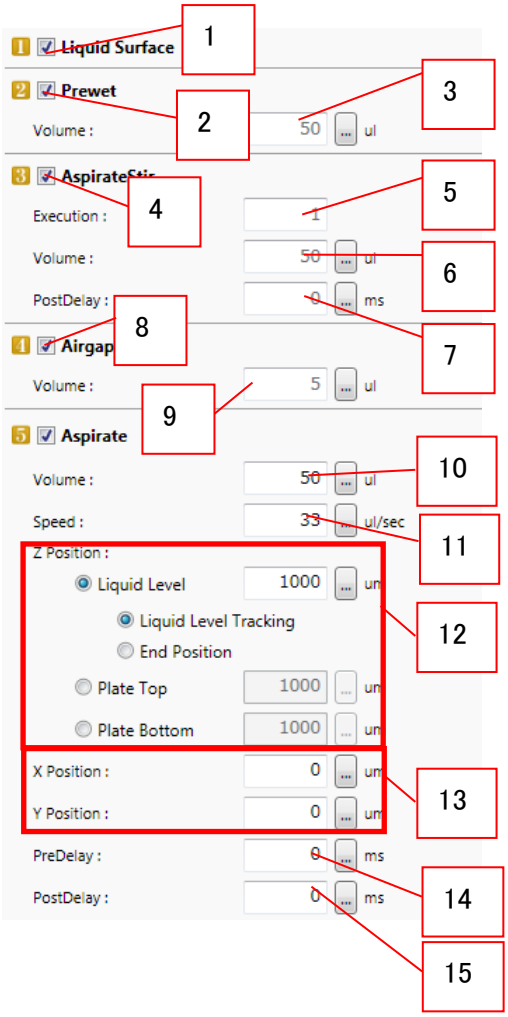


1) Liquid Surface function ON/OFF
2) Prewet function ON/OFF
3) Amount of solution suctioned in the Prewet mode
4) AspirateStir function ON/OFF
5) Number of times solution is stirred in the AspirateStir mode
6) Amount of solution suctioned in the AspirateStir mode
7) Sleep time after the syringe operation in the AspirateStir mode
8) Airgap function ON/OFF
9) Airgap volume
10) Amount of solution filled in the Aspirate mode
11) Filling speed in the Aspirate mode
12) Tip position at which solution is aspirated in the Aspirate mode
    Liquid Level: Track the drop in liquid level due to filling
    The value to be entered means distance between the tip and liquid level.
    Plate Bottom: Use the bottom face of the plate as a reference
13) Sleep time after the syringe operation in the Aspirate mode
14) AspirateTiptouch function ON/OFF

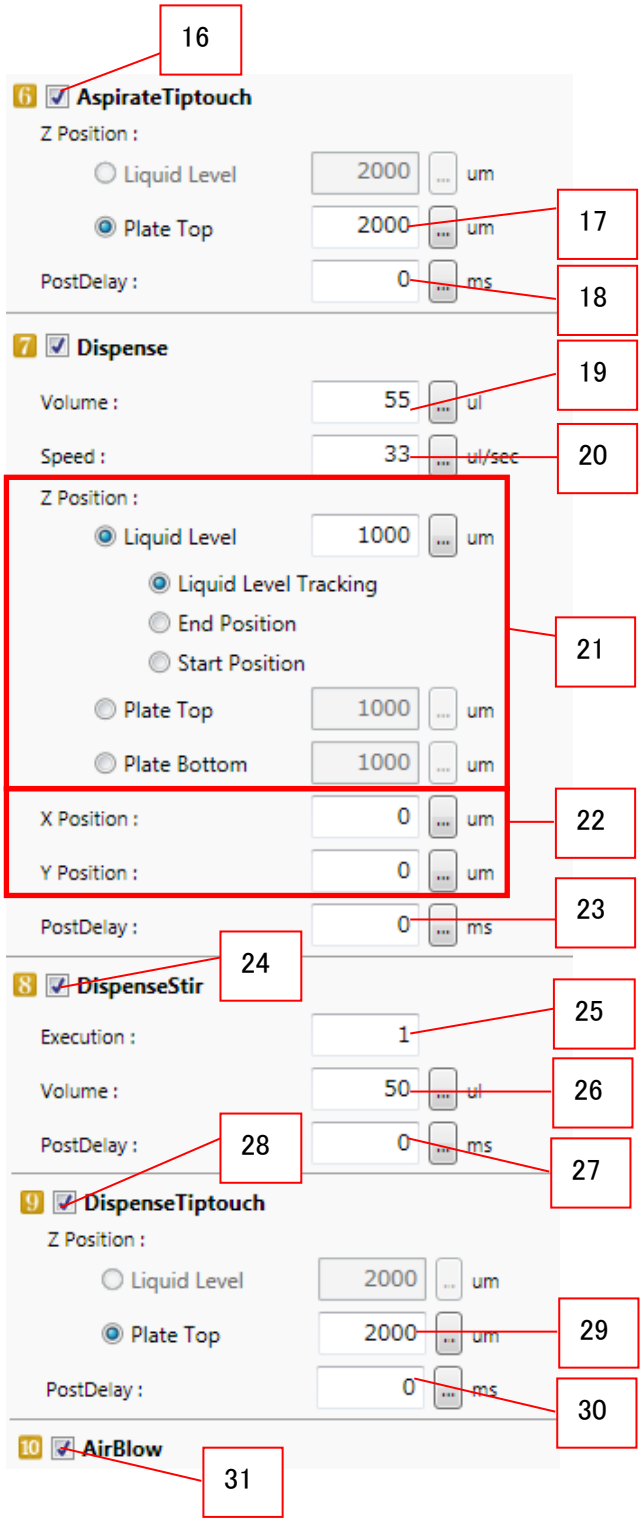


15) Amount of solution dripped in the Dispense mode
16) Dripping speed in the Dispense mode
17) Tip position at which solution is dripped in the Dispense mode
Liquid Level：Distance between the tip and liquid level
Liquid Level Tracking：Track the rise in liquid level due to dripping
End Position：Drip the solution from the liquid level after dripping
Start Position：Drip the solution from the liquid level before dripping
Plate Top：Use the top face of the plate as a reference
Plate Bottom: Use the bottom face of the plate as a reference
18) Sleep time after the syringe operation in the Dispense mode
19) DispenseStir function ON/OFF
20) Number of times solution is stirred in the DispenseStir mode
21) Amount of solution suctioned in the DispenseStir mode
22) Sleep time after the syringe operation in the DispenseStir mode
23) DispenseTiptouch function ON/OFF
24) AirBlow function ON/OFF

Advanced Setting Screen



1) Liquid Surface function ON/OFF
2) Prewet function ON/OFF
3) Amount of solution suctioned in the Prewet mode
4) AspirateStir function ON/OFF
5) Number of times solution is stirred in the AspirateStir mode
6) Amount of solution suctioned in the AspirateStir mode
7) Sleep time after the syringe operation in the AspirateStir mode
8) Airgap function ON/OFF
9) Airgap volume
10) Amount of solution filled in the Aspirate mode
11) Filling speed in the Aspirate mode
12) Z position of the tip when filling in the Aspirate mode
    Liquid Level：Distance between the tip and liquid level
    Liquid Level Tracking：Track the drop in liquid level due to filling
    End Position：Fill the solution from the liquid level after filling
    Plate Top：Use the top face of the plate as a reference
    Plate Bottom: Use the bottom face of the plate as a reference
13) XY positions of the tip when filling in the Aspirate mode
14) Sleep time before the syringe operation in the Aspirate mode
15) Sleep time after the syringe operation in the Aspirate mode

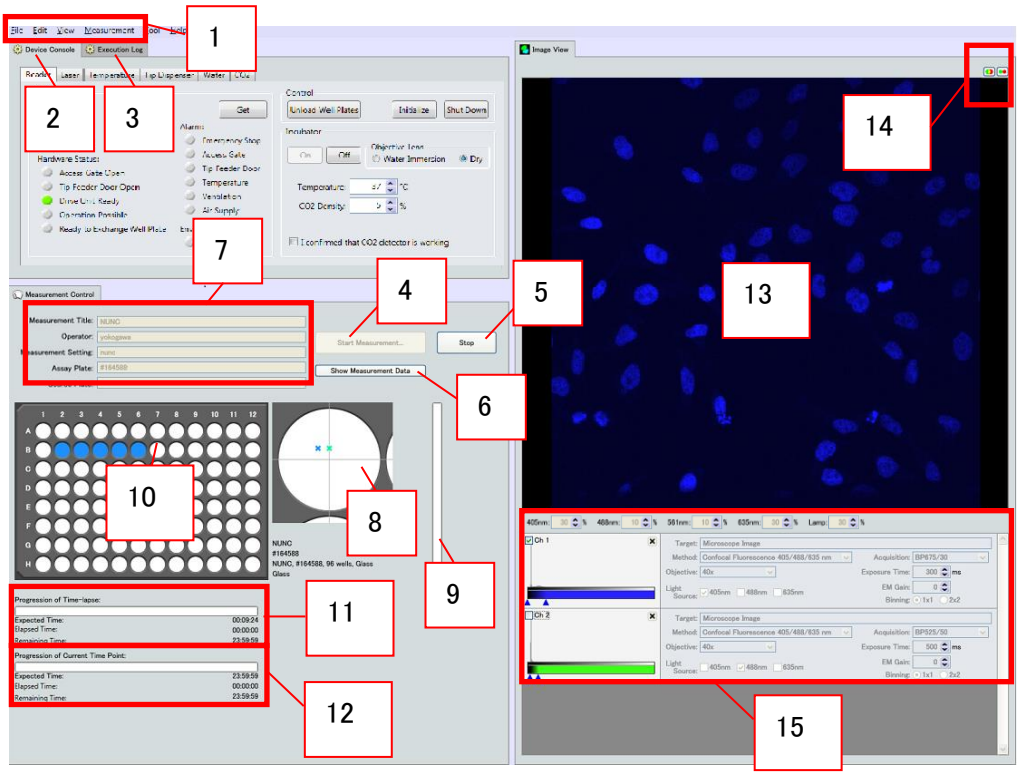


16) AspirateTiptouch function ON/OFF
17) Z position at which AspirateTiptouch is performed
Plate Top：Use the top face of the plate as a reference
18) Sleep time after the syringe operation in the AspirateTiptouch mode
19) Amount of solution dripped in the Dispense mode
20) Dripping speed in the Dispense mode
21) Tip position at which solution is dripped in the Dispense mode
Liquid Level：Distance between the tip and liquid level
Liquid Level Tracking：Track the rise in liquid level due to dripping

End Position：Drip the solution from the liquid level after dripping
Start Position：Drip the solution from the liquid level before dripping
Plate Top：Use the top face of the plate as a reference
Plate Bottom: Use the bottom face of the plate as a reference

22) XY positions of the tip when dripping in the Dispense mode
23) Sleep time after the syringe operation in the Dispense mode
24) DispenseStir function ON/OFF
25) Number of times solution is stirred in the DispenseStir mode
26) Amount of solution suctioned in the DispenseStir mode
27) Sleep time after the syringe operation in the DispenseStir mode
28) DispenseTiptouch function ON/OFF
29) Z position at which DispenseTiptouch is performed
Plate Top：Use the top face of the plate as a reference
30) Sleep time after the syringe operation in the DispenseTiptouch mode
31) AirBlow function ON/OFF

6.4. Reader Control Screen



1) Menus

File menu	Explanation
Open Well Plate Information List	Open the screen for selecting a well plate information file.
Open Measurement Setting List	Open the screen for selecting a measurement setting file.
Open Dispense Setting List	Open the screen for selecting a dispensing setting file.
Open Measurement Data List	Open the screen for selecting measured results.
Register User Well Plate	Register new well plate information.
Edit User Well Plate	Edit well plate information registered by user.
Delete User Well Plate	Delete well plate information registered by user.
Import User Well Plate	Import well plate information.
Export User Well Plate	Export well plate information registered by user.
Open Fluorophore Manager	Open the screen for registering fluorophore.
Open Log List	Open Log List screen for temperature and CO2.
Close	Close the Reader Control screen.

Edit menu	Explanation
Undo	Undo the last operation.
Redo	Redo the last operation.
Cut	Cut the selected item.
Copy	Copy the selected item.
Paste	Paste the selected item.

View menu	Explanation
Device Console	Open the Device Console tab.
Execution Log	Open the Execution Log tab.
Show Overlay Images	Show overlaid images of multiple channels.
Show Tile Images	Show images for each channel.

Measurement menu	Explanation
Start Measurement	Start measurement.
Stop Measurement	Stop measurement.

Tool menu	Explanation
Default Image Correction Setting for Automation	Show image correction in external control mode setting window

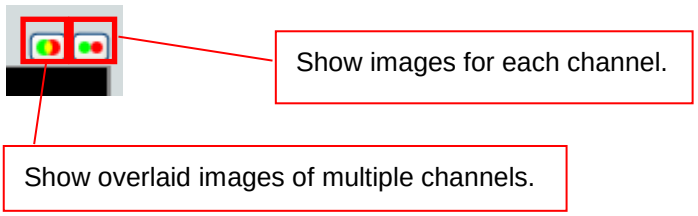
Help menu	Explanation
About	Show the version information of the measurement software.

2) Open the screen for setting the laser, heater, etc.
3) Open the log screen.
4) Start measurement.
5) Stop measurement.
6) Show measured results.
7) Information items entered at the time of measurement

Item	Explanation
Measurement Title	Title of measurement
Operator	Name of the person who performed measurement
Measurement Setting	Measurement setting file
Assay Plate	Assay Plate name
Source Plate	Source Plate name

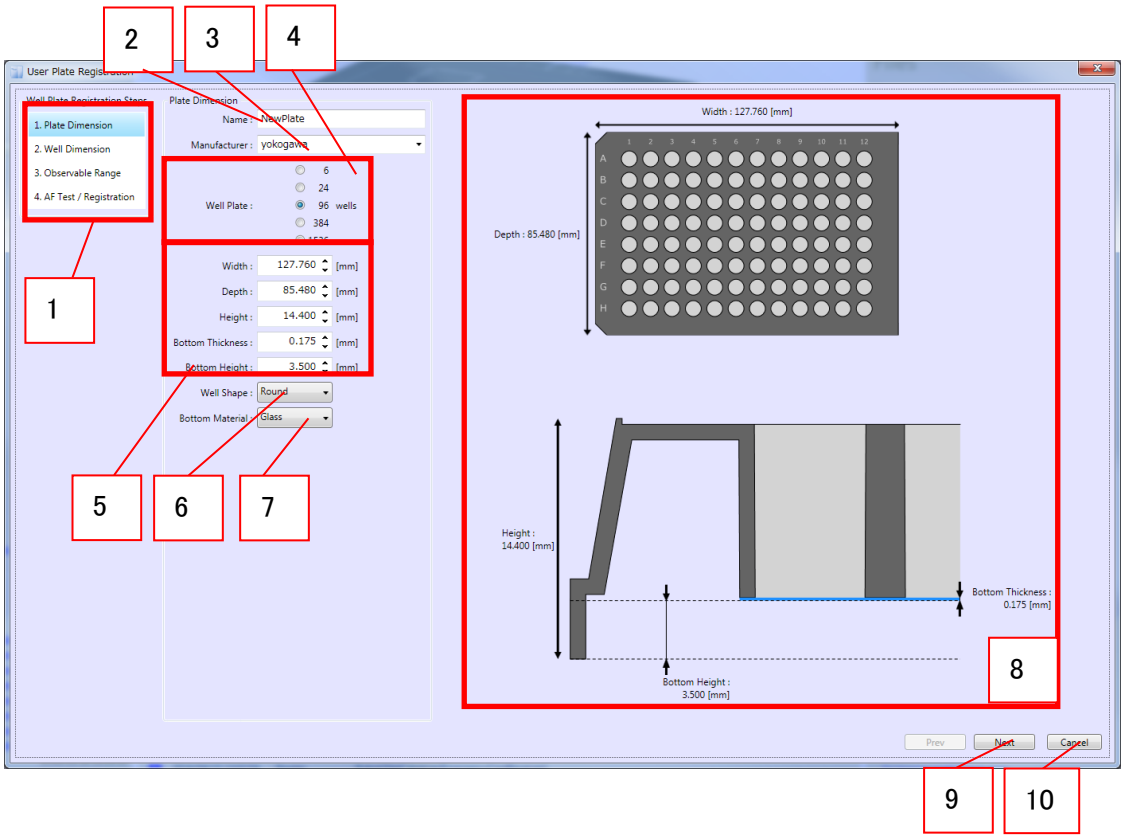
8) View field of measurement
9) Progress bar for measurement in Z-axis direction
10) The progress of measurement is indicated by well plates.
11) Progress status display of time-lapse measurement
Expected Time：Expected time at which all measurement will end
Current Time：Elapsed time after start of measurement
Remaining Time：Remaining time to end of measurement
12) Progress status display of processing at one time point
Expected Time：Time at which measurement at the time point will end
Current Time：Elapsed time after start of measurement at the time point
Remaining Time：Remaining time to end of measurement at the time point
13) Image currently being captured

14) Select the display format for channel images.



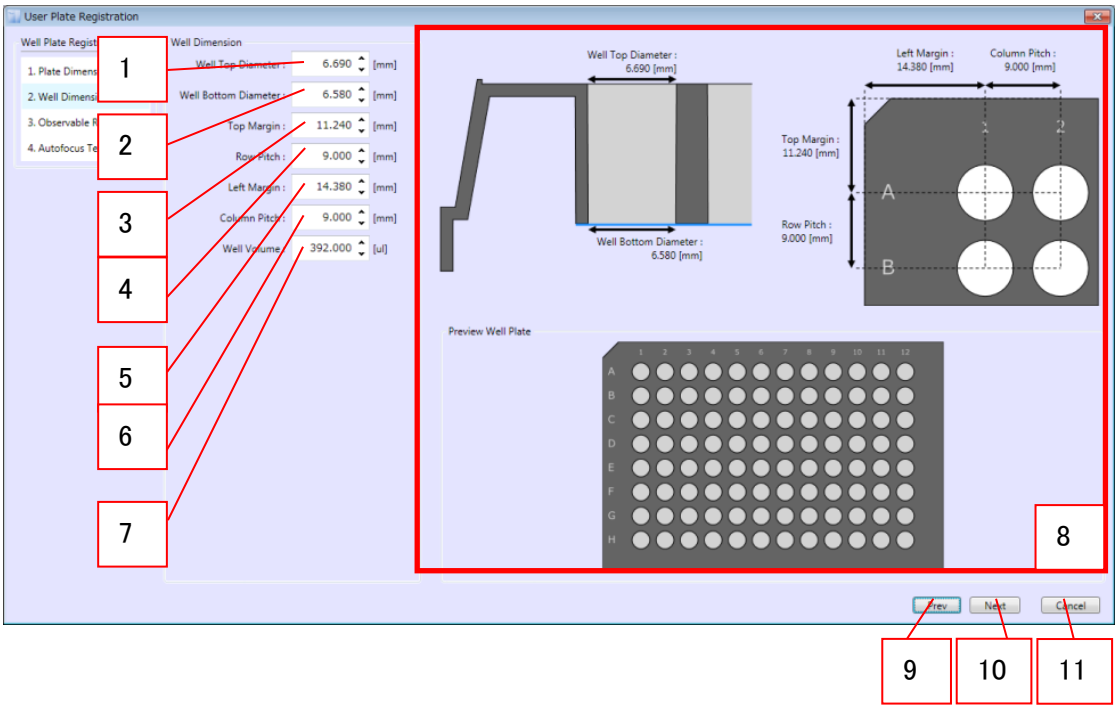
15) The channel settings and laser output are shown.

User Plate Registration Screen (Plate Dimension)



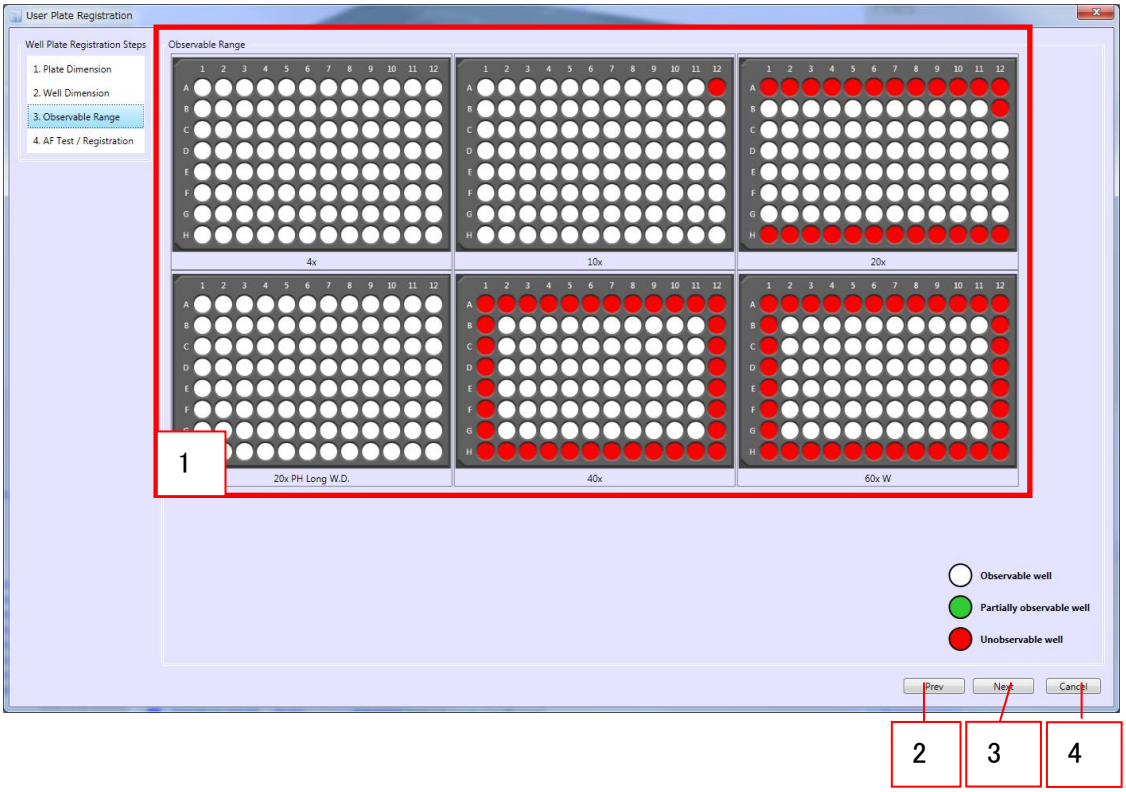
1) Step of registration of well plate information.
2) Name of well plate.
3) Manufacturer of well plate.
4) Number of wells.
5) Dimension of well plate.
6) Shape of well.
7) Bottom material of well plate.
8) Showing dimension and shape of well plate
9) Go to next step with saving changes.
10) Close the window without saving changes.

User Plate Registration Screen (Well Dimension)



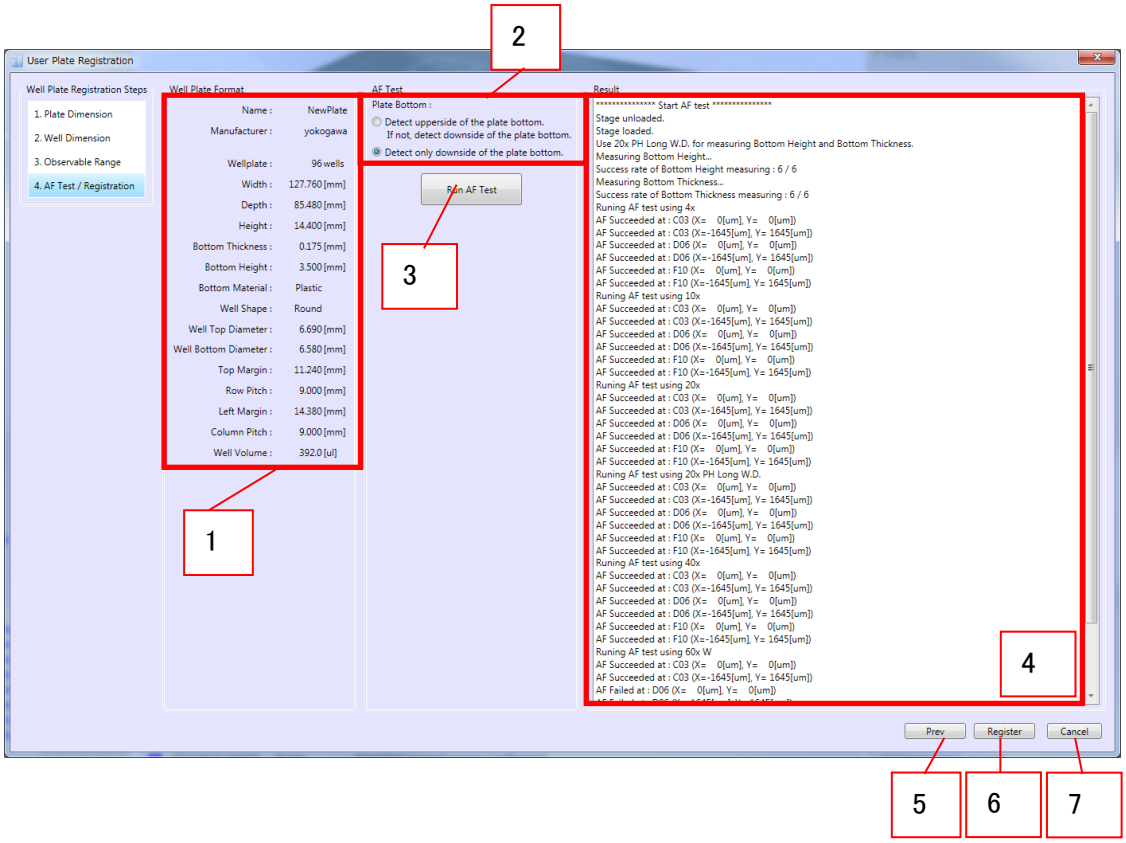
1) Diameter (round well)/ side length (rectangle well) of well top.
2) Diameter (round well)/ side length (rectangle well) of well bottom.
3) Distance between plate upper end and center of the first row well.
4) Row pitch of wells
5) Distance between plate left end and center of the first column well.
6) Column pitch of wells.
7) Volume of well.
8) Showing dimension and shape of well.
9) Return to previous step with saving changes.
10) Go to next step with saving changes.
11) Close the window without saving changes.

User Plate Registration Screen (Observable Range)



1) Showing observable range of each objective lens.
   White: Whole well is observable
   Green: Partial of well is observable
   Red: Whole well is unobservable
2) Return to previous step.
3) Go to next step.
4) Close the window without saving changes.

User Plate Registration Screen (Autofocus Test)



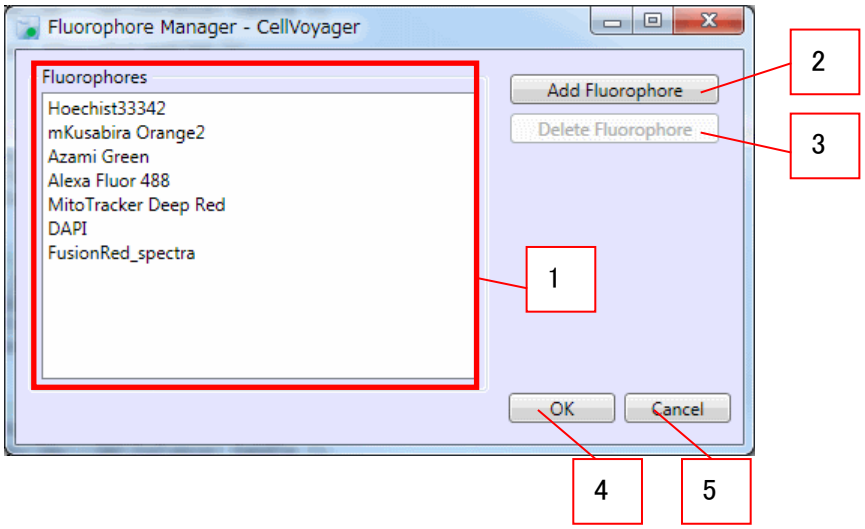
1) Well plate information.

2) Setting of auto focus detecting plane.

	Explanation
Detect upperside of the plate bottom. If not, detect downside of the plate bottom.	Perform auto focus test on upper side of the plate bottom at first. If succeed, it is set that auto focus is performed on upper side of the plate bottom in this plate. In case of auto focus on upper side of the plate bottom fails, auto focus test on lower side of the plate bottom is performed. If succeed, it is set that auto focus is performed on lower side of the plate bottom in this plate. In case of auto focus on both sides of the plate bottom fails, information of this plate is not registered.
Detect only downside of the plate bottom.	Perform auto focus test on only lower side of the plate bottom at. If succeed, it is set that auto focus is performed on lower side of the plate bottom in this plate. In case of auto focus on lower side of the plate bottom fails, information of this plate is not registered.

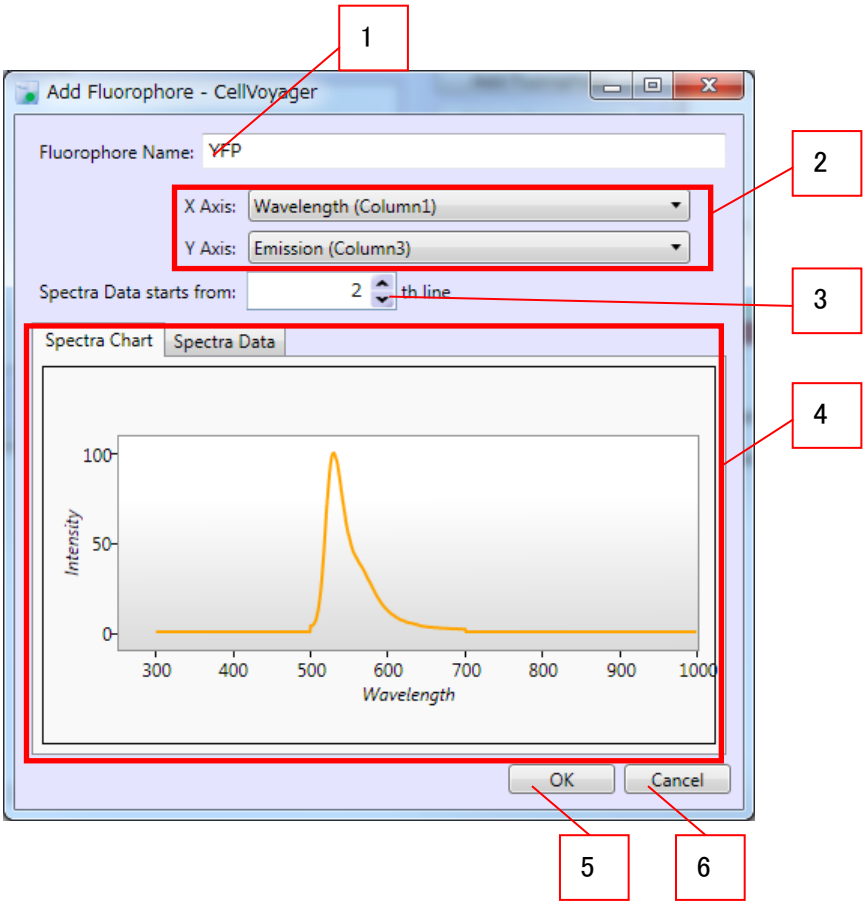
3) Start auto focus test.
4) Result of auto focus test.
5) Return to previous step.
6) Close the window with registering plate information.
7) Close the window without registering plate information.

Fluorophore Manager Screen



1) List of registered fluorophore.
2) Add fluorophore
3) Delete fluorophore.
4) Close the window with saving changes.
5) Close the window without saving changes.

Add Fluorophore Screen

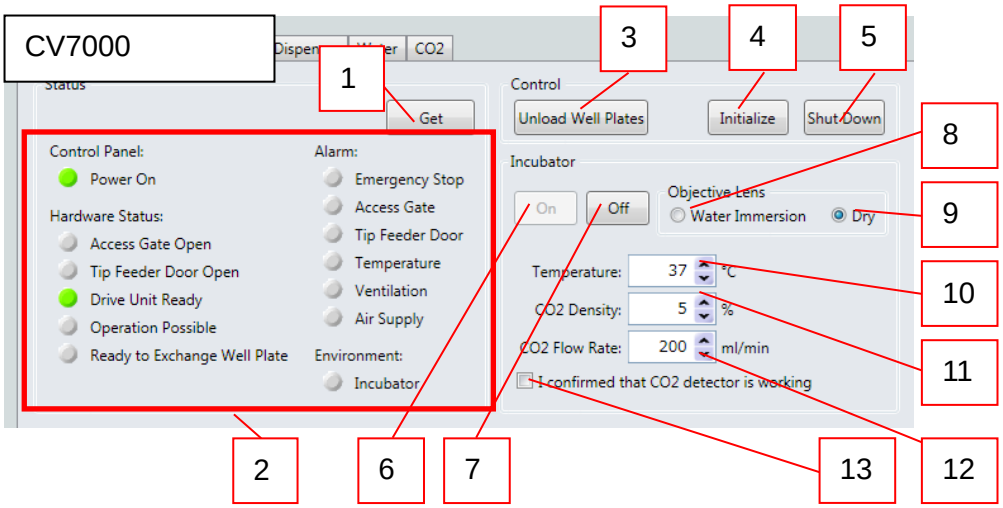


1) Fluorophore name.
2) Select wavelength data column and emission spectral data column.
3) Select line that wavelength data and emission spectral data start.
4) Display fluorophore spectral data.
   Spectra Chart: Display graph
   Spectra Data: Display numeric data
5) Close the window with determining fluorophore spectral data.
6) Close the window without determining fluorophore spectral data.

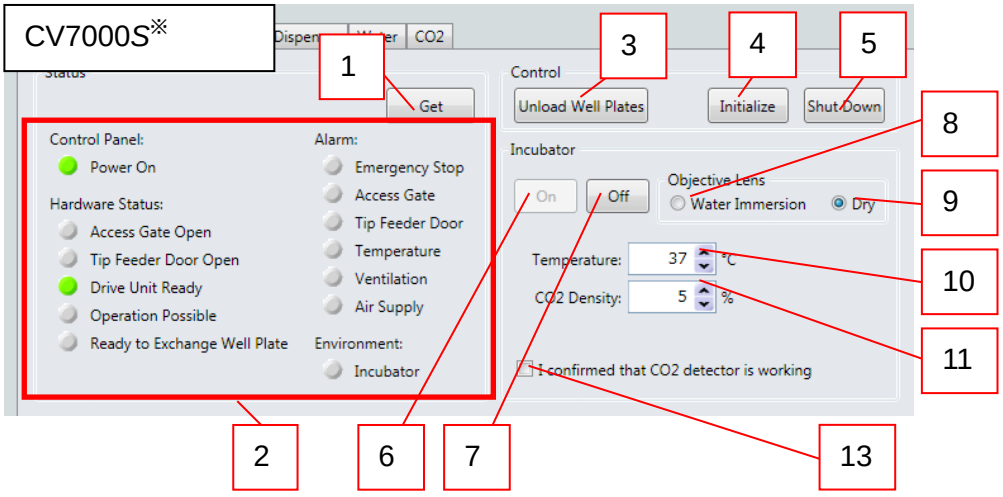
Device Console Screen

Reader tab

CV7000



CV7000S※



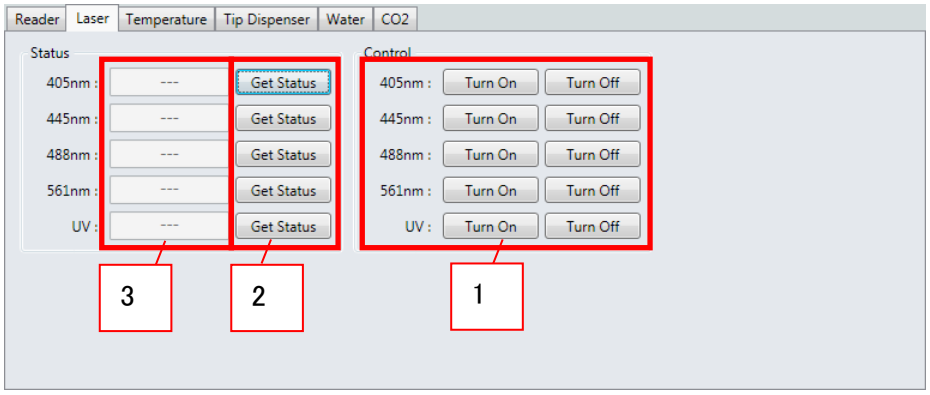
1) Acquire status information.
2) Status information

Status information		Explanation
Control Panel	Power On	The power is on.
Hardware Status	Access Gate Open	The access gate is open.
	Tip Feeder Door Open	The tip platform gate is open.
	Drive Unit Ready	The device unit is ready.
	Operation Possible	Accessible from the measurement software.
	Ready to Exchange Well Plate	Well plates can be exchanged.
Alarm	Emergency Stop	An emergency stop has been actuated.
	Access Gate	An access gate error has occurred.
	Tip Feeder Door	A tip platform gate error has occurred.
	Temperature	A temperature error has occurred.
	Internal Environment	An internal environment fan error has occurred.
	Air Supply	An air cylinder error has occurred.
Environment	Incubator	The incubator becomes stable.

3) Move the stage to in front of the access gate on the front side of the measurement section.
4) Start/restart the system.
5) Shut down the system.
6) Turn on the stage incubator.
7) Turn off the stage incubator.
8) Select the water immersion lenses.
9) Select the dry lenses.
10) Enter the heater temperature
11) Enter the value of CO_2 concentration.
12) Enter the value of gas flow rate to the stage incubator.
13) Check box for confirmation that CO_2 detector is working.
(If unchecked, CO_2 supply doesn't start.)

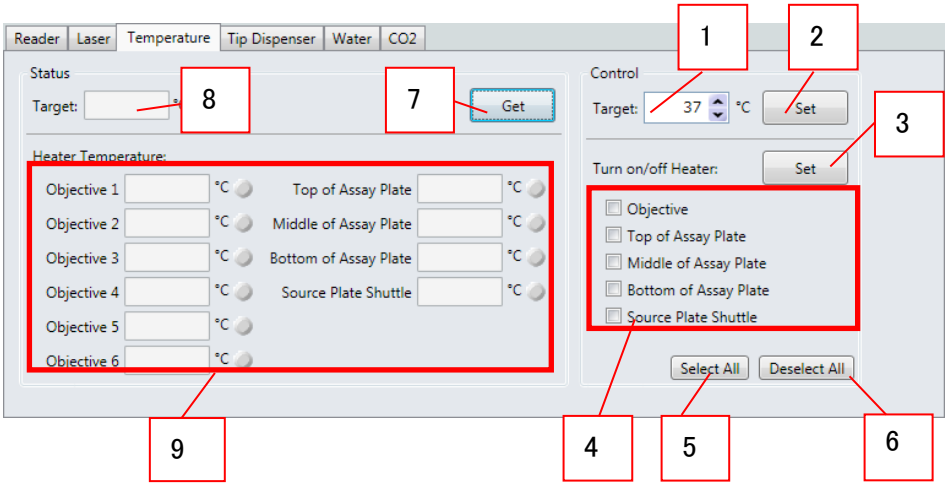
※ Gas flow rate can't be set in CV7000*S*, which is sold after April 2015. (Refer to 3.4.)

Laser Tab



1) Start/shut down the laser (Turn On/Turn Off)
   405nm：405 nm wavelength laser
   488nm：488 nm wavelength laser
   561nm(or 532nm)：561 nm(or 532nm) wavelength laser
   640nm：640 nm wavelength laser
2) Acquire laser status information.
3) Laser status information is shown.
   Turn On：Laser is already running.
   Turn Off：Laser is off.
   Starting：Laser is starting.

Temperature Tab



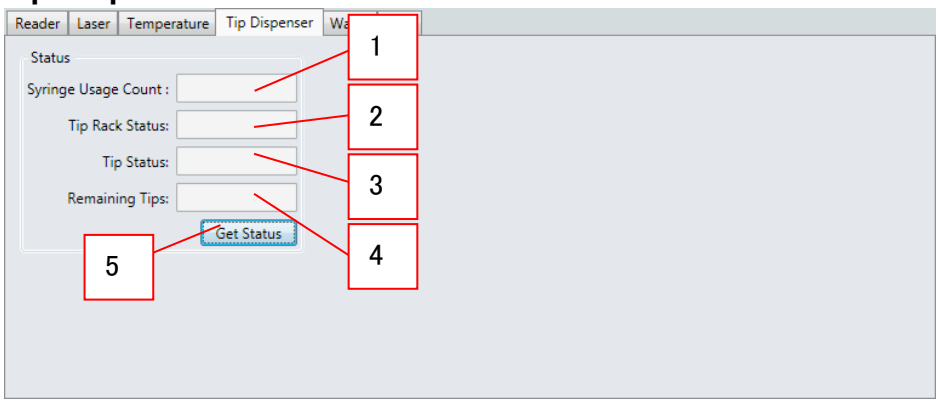
1) Set heater temperature
2) Reflect the set heater temperature.
3) Turn on the heater for the selected targets of temperature control.
4) Select the targets of temperature control.

Temperature setting location	Explanation
Objective	Object lens heater
Top of Assay Plate	Assay plate top stage heater
Middle of Assay Plate	Assay plate center stage heater
Bottom of Assay Plate	Assay plate bottom stage heater
Source Plate Shuttle	Source plate heater

5) Select all as targets of temperature control.
6) Unselect all.
7) Acquire status information.
8) Set temperature
9) Temperature conditions

Item	Explanation
Objective 1	The temperature of the first positional object lens
Objective 2	The temperature of the second positional object lens
Objective 3	The temperature of the third positional object lens
Objective 4	The temperature of the fourth positional object lens
Objective 5	The temperature of the fifth positional object lens
Objective 6	The temperature of the sixth positional object lens
Top of Assay Plate	Assay plate top stage temperature
Middle of Assay Plate	Assay plate center stage temperature
Bottom of Assay Plate	Assay plate bottom stage temperature
Source Plate Shuttle	Source plate stage temperature

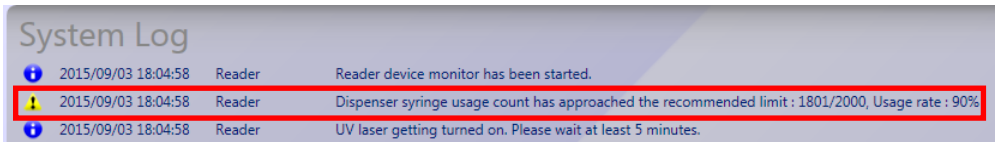
Tip Dispenser tab



1) Number of times that dispenser syringe is used.

MEMO

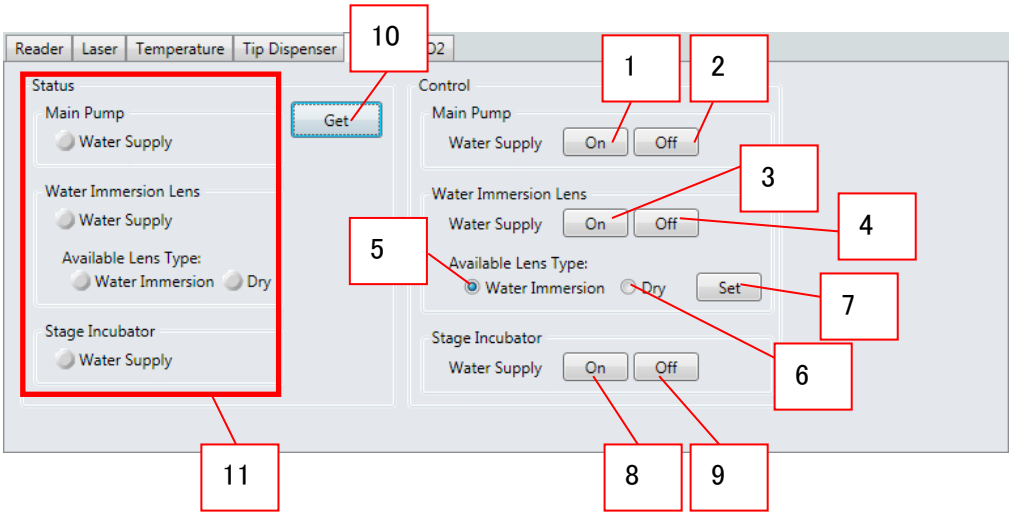
●After number of times that syringe is used reach 80 % of operating life, caution is displayed on “System Log”.



It is recommended to exchange syringe when this caution appears. Please contact us.

2) Whether or not there is a tip rack at the dispenser (Ready/Not Ready/Not Available)
3) Tip setting condition at the dispenser (Attached/Not Attached)
4) Number of tips remaining on the tip rack at the dispenser (96-tip rack model: 0 to 96) (384-tip rack model: 0 to 384)
5) Acquire status information.

Water tab

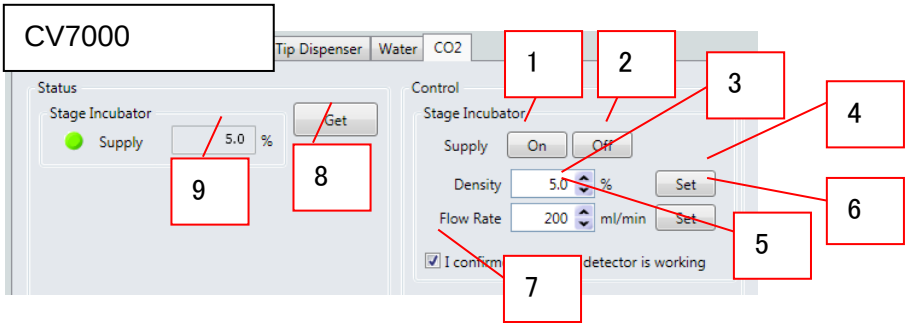


1) The water is supplied to the main pump.
2) The water is discharged from the main pump.
3) The water is supplied to the water immersion lenses.
4) The water is not supplied to the water immersion lenses.
5) Select the water immersion lenses.
6) Select the dry lenses.
7) Set the selected lens type.
8) The water is supplied to the stage incubator.
9) The water is not supplied to the stage incubator.
10) Acquire status information.
11) Status information is shown.

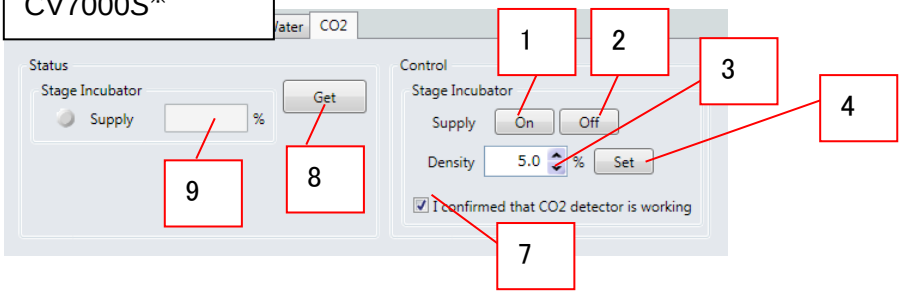
Status information		Explanation
Main Pump	Water Supply	Water supply to the main pump has been executed.
Water Immersion Lens	Water Supply	Water supply to the water immersion lenses has been executed.
Available Lens Type	Water Immersion	Water Immersion lenses can be used.
	Dry	Dry lenses can be used.
Stage Incubator	Water Supply	Water supply to the stage incubator has been executed.

CO_2 tab

CV7000



CV7000S※



1) CO_2 is supplied to the stage incubator.
2) CO_2 is not supplied to the stage incubator.
3) Enter the value of CO_2 concentration.
4) Set the entered CO_2 concentration.
5) Enter the value of gas flow rate to the stage incubator.
6) Set the value of gas flow rate.
7) Check box for confirmation that CO_2 detector is working.
   (If unchecked, CO_2 supply doesn't start.)
8) Acquire status information.
9) Status information for CO_2 concentration to be supplied.

※ Gas flow rate can't be set in CV7000S, which is sold after April 2015. (Refer to 3.4.)

7. Setting Examples of Measurement Setting Files

7.1. Imaging by Auto-focus

Images are captured without using the software focus.

Confocal Imaging by Auto-focus

Confocal imaging is performed on the auto-focus plane at two wavelengths (405 nm, 488 nm).

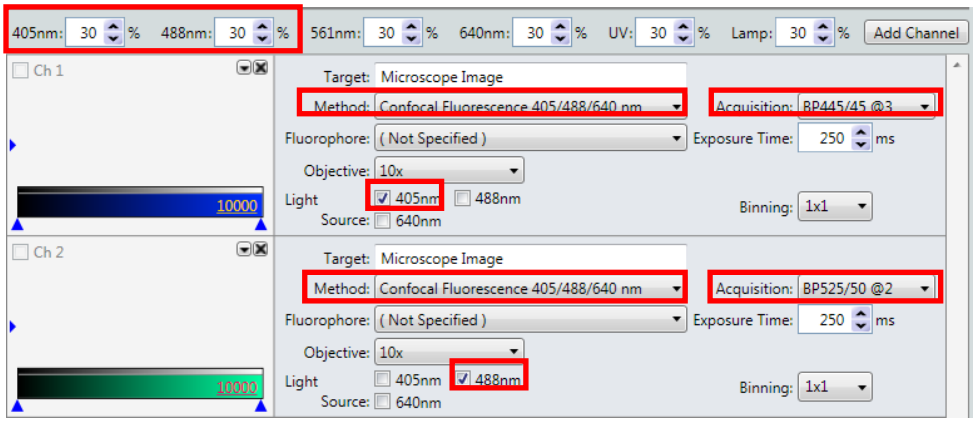
1) Open the measurement setting file edit screen. (Refer to 5.2)

2) Enter the application name. (Refer to 5.4)

3) Set the imaging channels. (Refer to 5.5)

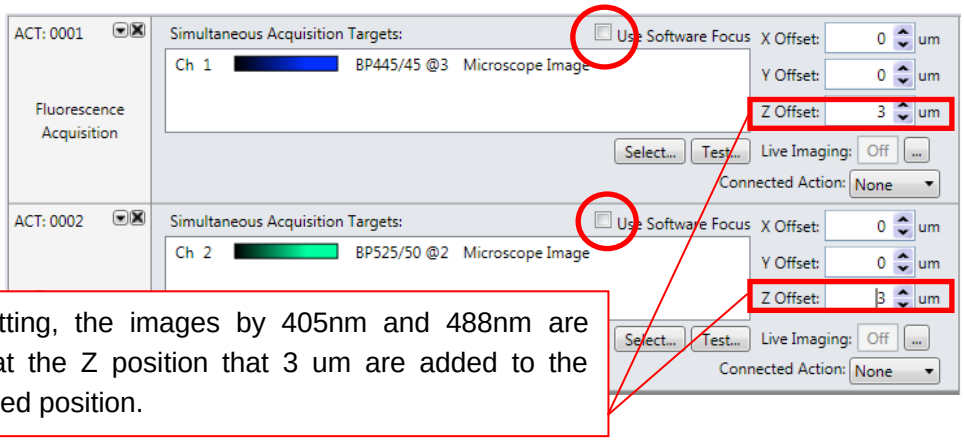
Ch1 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP445/45
Light Source : 405nm (30%)

Ch2 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP525/50
Light Source : 488nm (30%)



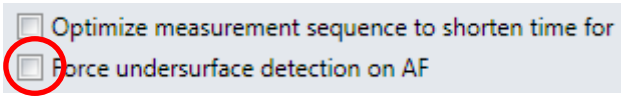
4) Set the items on the Action List tab. (Refer to 5.7)

Set Ch1 and Ch2 for the two Fluorescence Acquisition tasks, respectively.
Unselect the “Use Software Focus” check box.



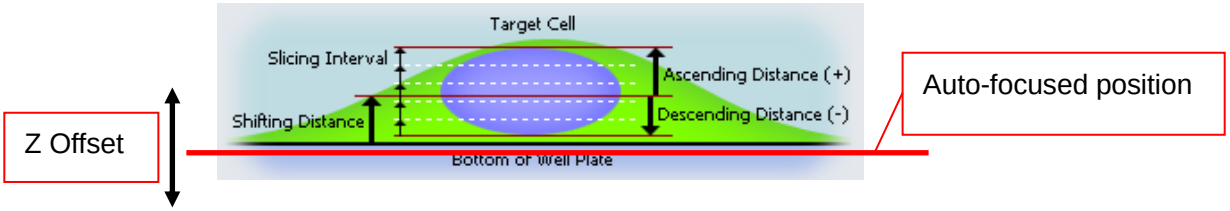
In this setting, the images by 405nm and 488nm are acquired at the Z position that 3 um are added to the auto-focused position.

Uncheck “Force undersurface detection on AF”.

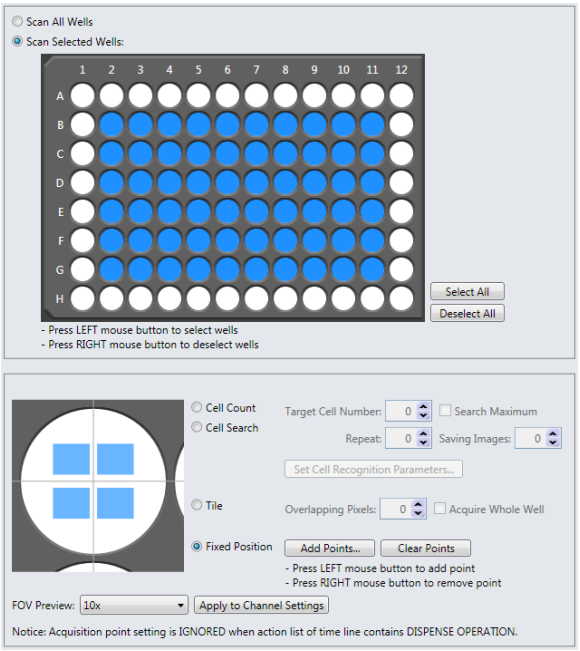


MEMO

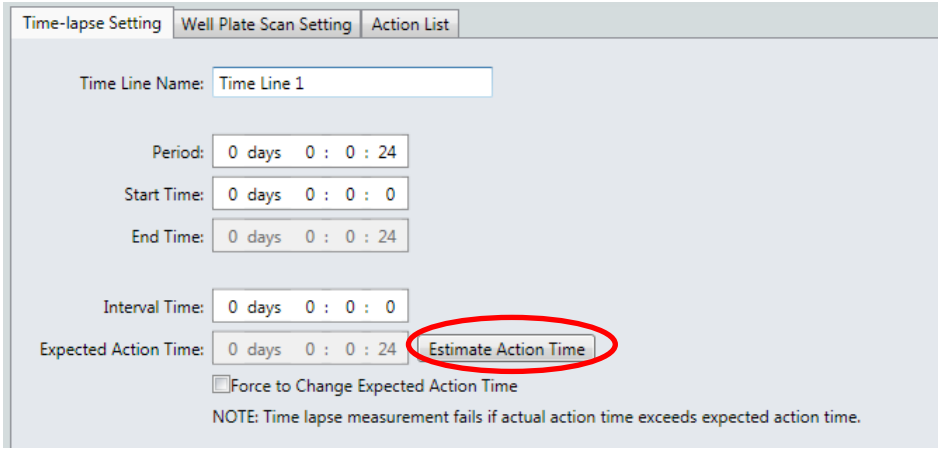
●Auto focus detects interface between medium and glass by the auto focus unit.



5) Set the imaging wells and view field on the Well Plate Scan Setting tab. (Refer to 5.6)



6) Click “Estimate Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3)



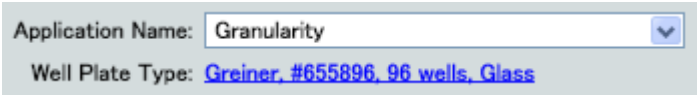
7) Save the measurement setting file. (Refer to 5.11)

Bright Field Imaging by Auto-focus

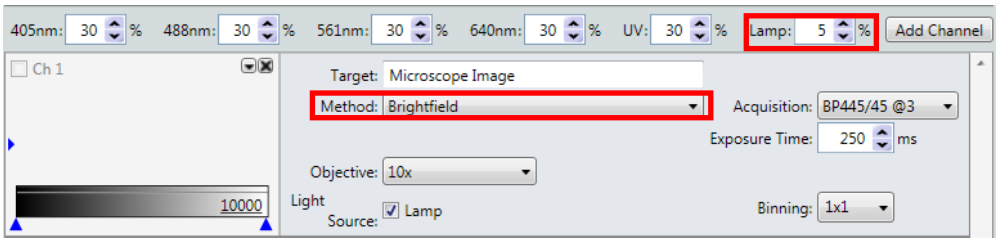
Bright field imaging is performed on the auto-focus plane.

1) Open the measurement setting file edit screen. (Refer to 5.2)

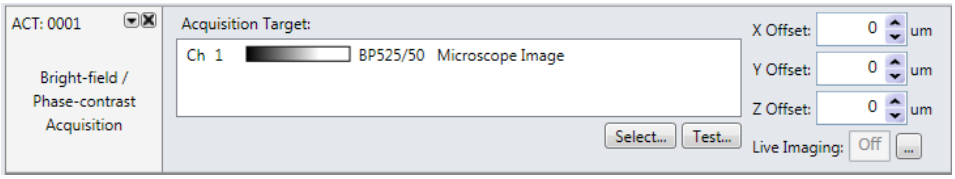
2) Enter the application name. (Refer to 5.4)



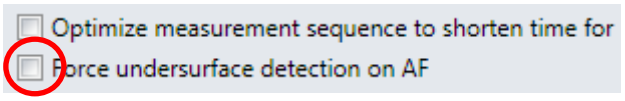
3) Set the imaging channels. (Refer to 5.5)
Ch1 Method : Brightfield
Light Source : Lamp (5%)



4) Set the items on the Action List tab. (Refer to 5.7)
Set Ch1 for the Bright-field/Phase-contrast Acquisition task.

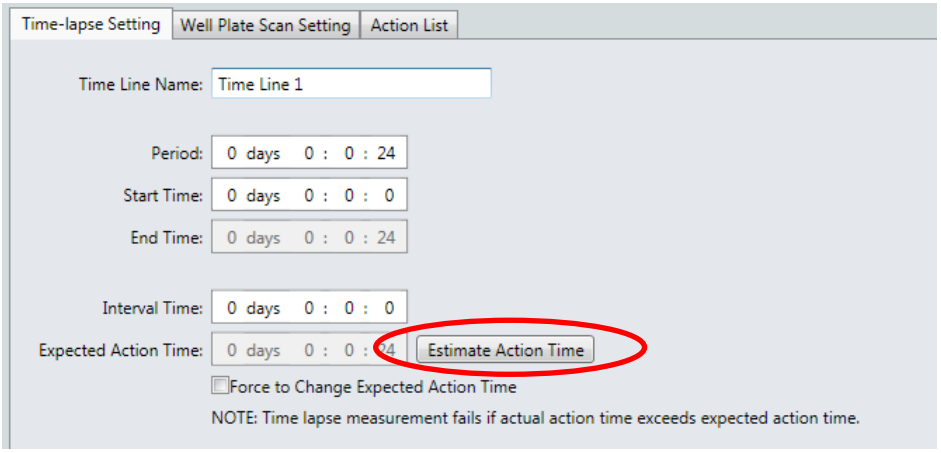


Uncheck “Force undersurface detection on AF”.



5) Set the imaging wells and view field on the Well Plate Scan Setting tab. (Refer to 5.6.)

6) Click “Estimate Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3)



7) Save the measurement setting file. (Refer to 5.11)

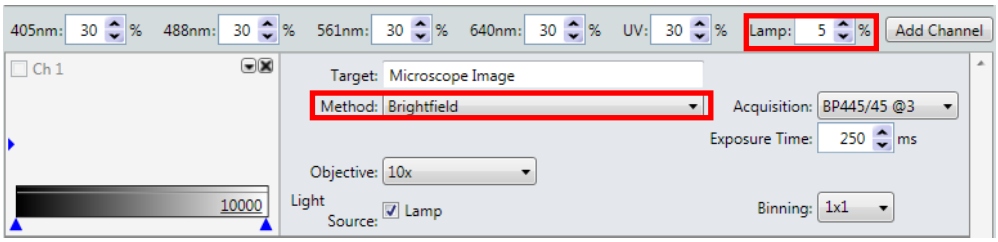
Z-Stack Imaging by Bright Field

Z-stack imaging by bright field is performed around the auto-focus plane.

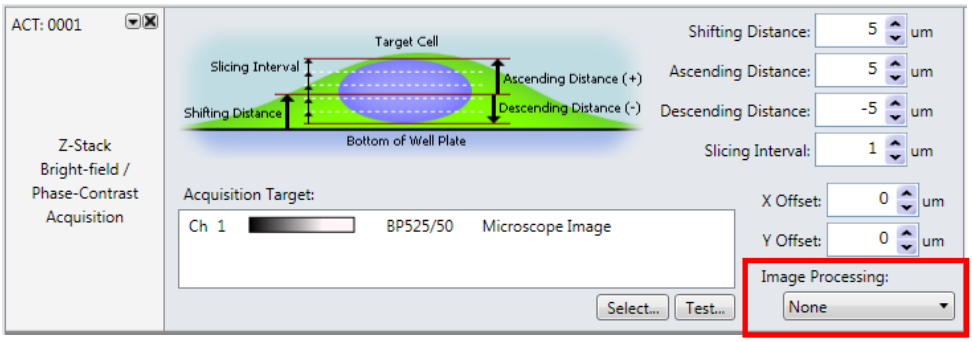
1) Open the measurement setting file edit screen. (Refer to 5.2)

2) Enter the application name. (Refer to 5.4)

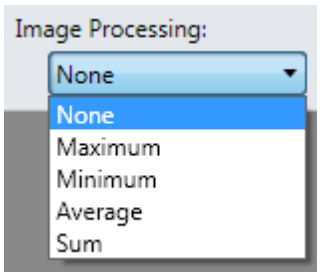
3) Set the imaging channels. (Refer to 5.5)
Ch1 Method : Brightfield
Light Source : Lamp (5%)



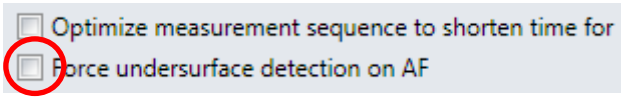
4) Set the items on the Action List tab. (Refer to 5.7)
Set Ch1 to Z-Stack Bright-field/Phase-Contrast Acquisition



Select an output method for Z images from “Image Processing.”
(Refer to 5.1 and 6.2.)

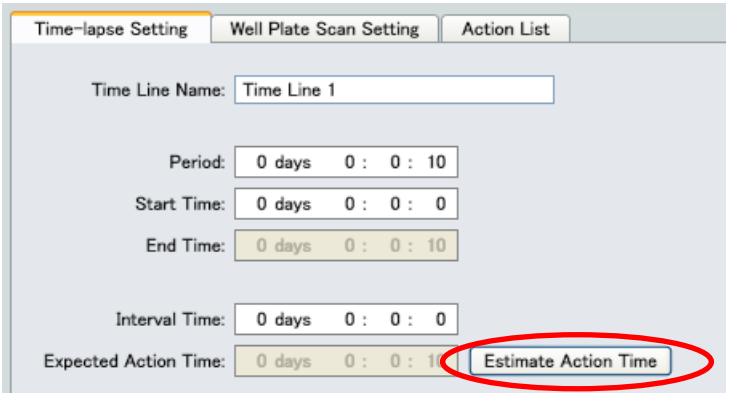


Uncheck “Force undersurface detection on AF”.



5) Set the imaging wells and view field on the Well Plate Scan Setting tab. (Refer to 5.6.)

6) Click “Estimate Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3)



7) Save the measurement setting file. (Refer to 5.11)

3D Imaging from the Auto-focus Plane

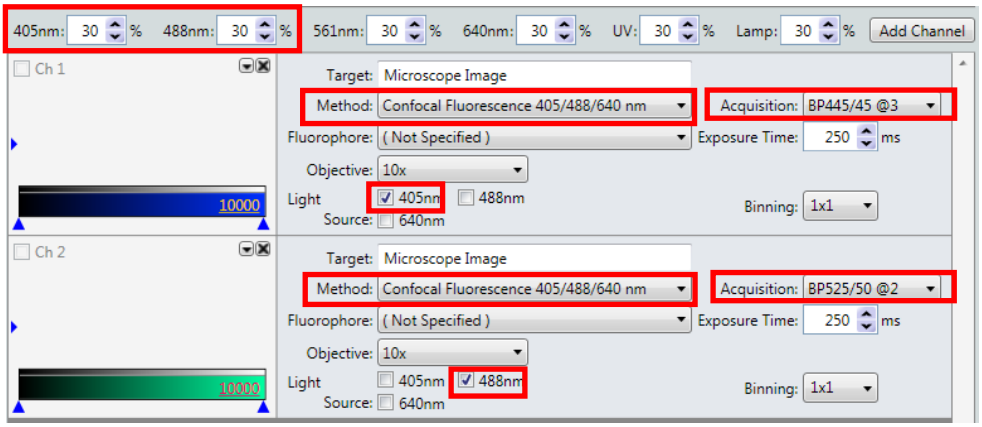
3D imaging is performed at two wavelengths (405 nm, 488 nm) by using the auto-focus plane as the reference.

1) Open the measurement setting file edit screen. (Refer to 5.2)

2) Enter the application name. (Refer to 5.4)

3) Set the imaging channels. (Refer to 5.5)

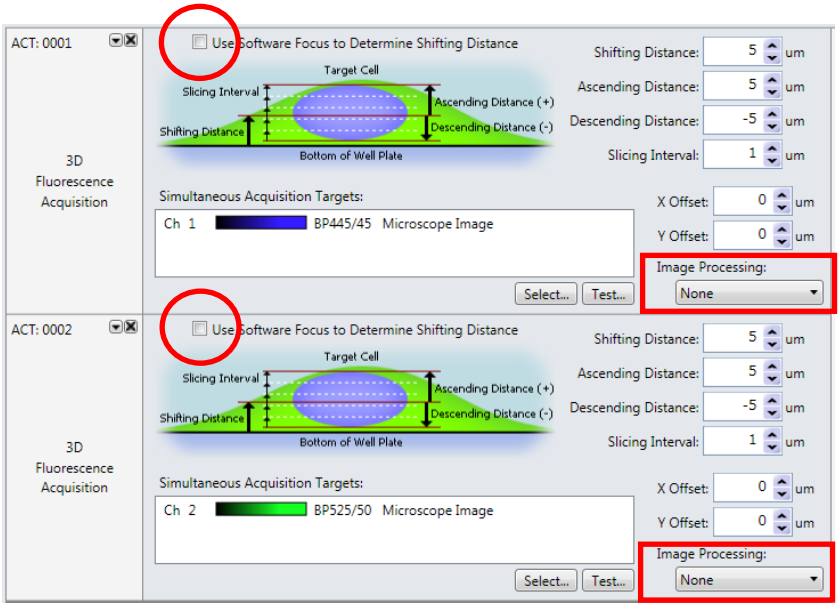
Ch1 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP445/45 Light Source : 405nm (30%)
Ch2 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP525/50 Light Source : 488nm (30%)



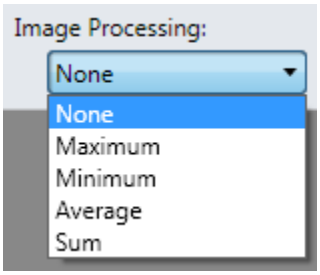
4) Set the items on the Action List tab. (Refer to 5.7)

Set Ch1 and Ch2 for the two 3D Fluorescence Acquisition tasks, respectively.

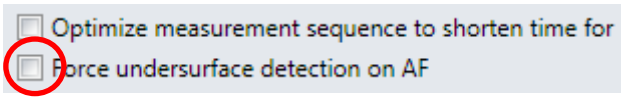
Unselect the “Use Software Focus to Determine Shifting Distance” check box.



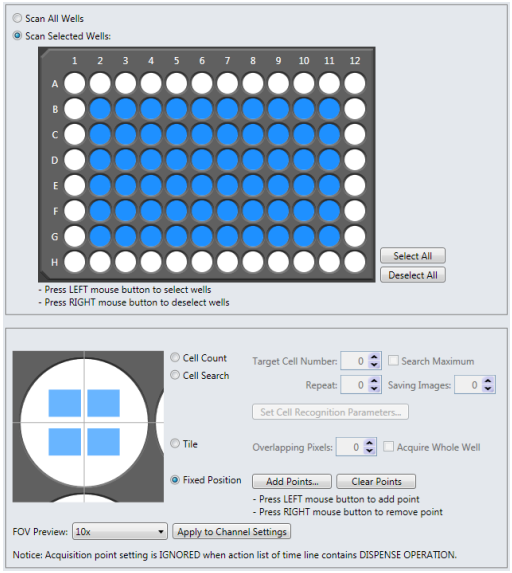
Select an output method for Z images from “Image Processing.”
(Refer to 5.1 and 6.2.)



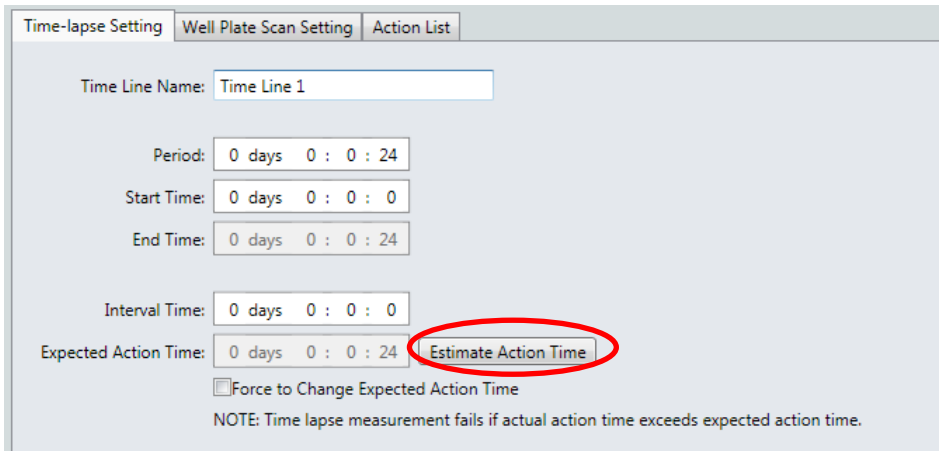
Uncheck “Force undersurface detection on AF”.



5) Set the imaging wells and view field on the Well Plate Scan Setting tab.
(Refer to 5.6)



6) Click “Estimate Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3)



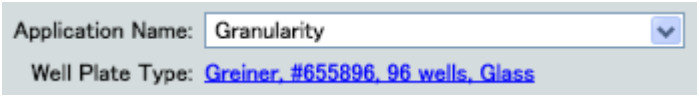
7) Save the measurement setting file. (Refer to 5.11)

Epifluorescence Imaging by Auto-focus

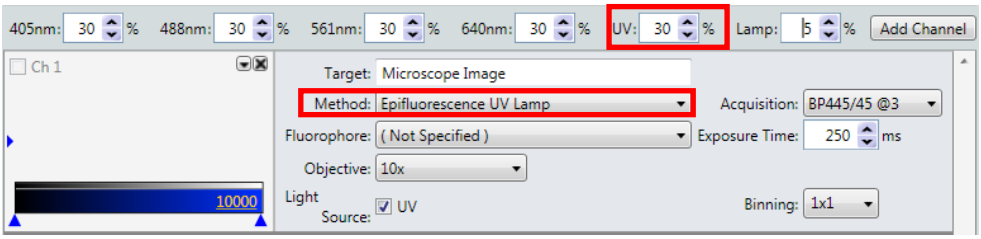
Epifluorescence imaging is performed on the auto-focus plane.

1) Open the measurement setting file edit screen. (Refer to 5.2)

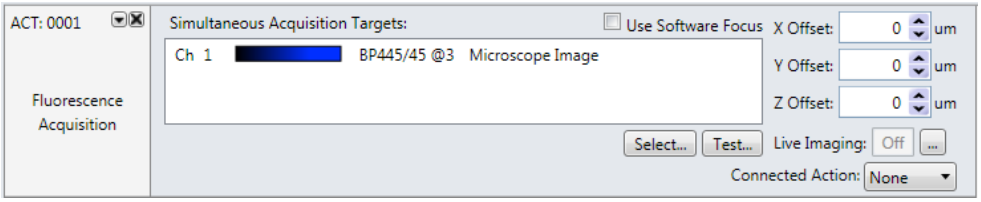
2) Enter the application name. (Refer to 5.4)



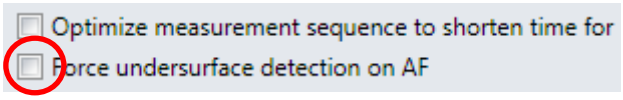
3) Set the imaging channels. (Refer to 5.5)
Ch1 Method : Epifluorescence UV Lamp (UV model only)
Light Source : UV (30%)



4) Set the items on the Action List tab. (Refer to 5.7)
Set Ch1 for the Fluorescence Acquisition task.

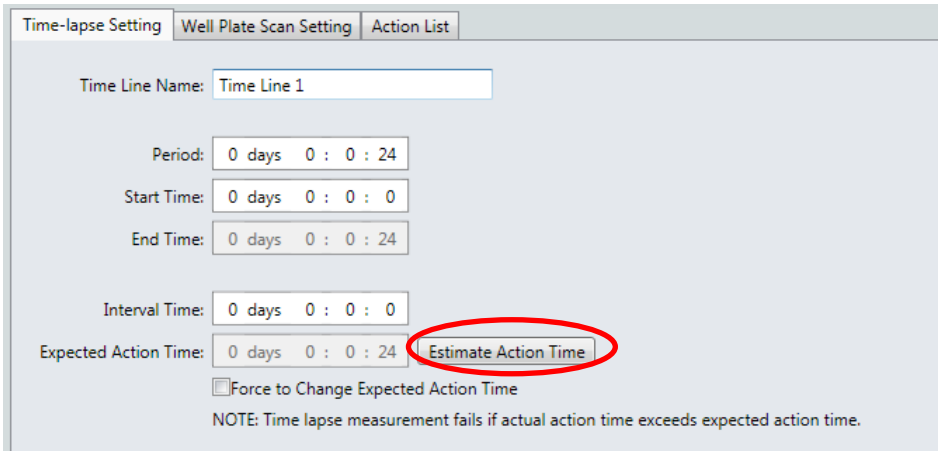


Uncheck “Force undersurface detection on AF”.



5) Set the imaging wells and view field on the Well Plate Scan Setting tab.
(Refer to 5.6)

6) Click “Estimate Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3)



7) Save the measurement setting file. (Refer to 5.11)

7.2. Imaging by Software Focus

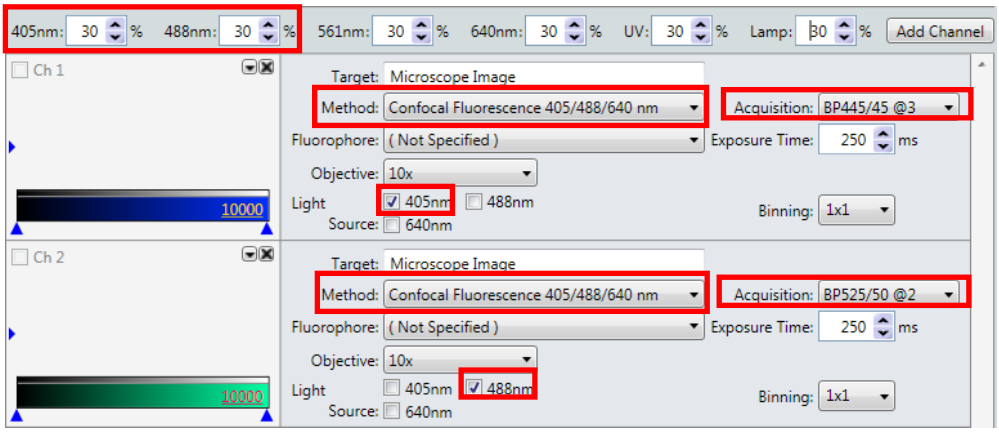
Imaging is performed on the software focus plane.

Confocal Imaging by Software Focus

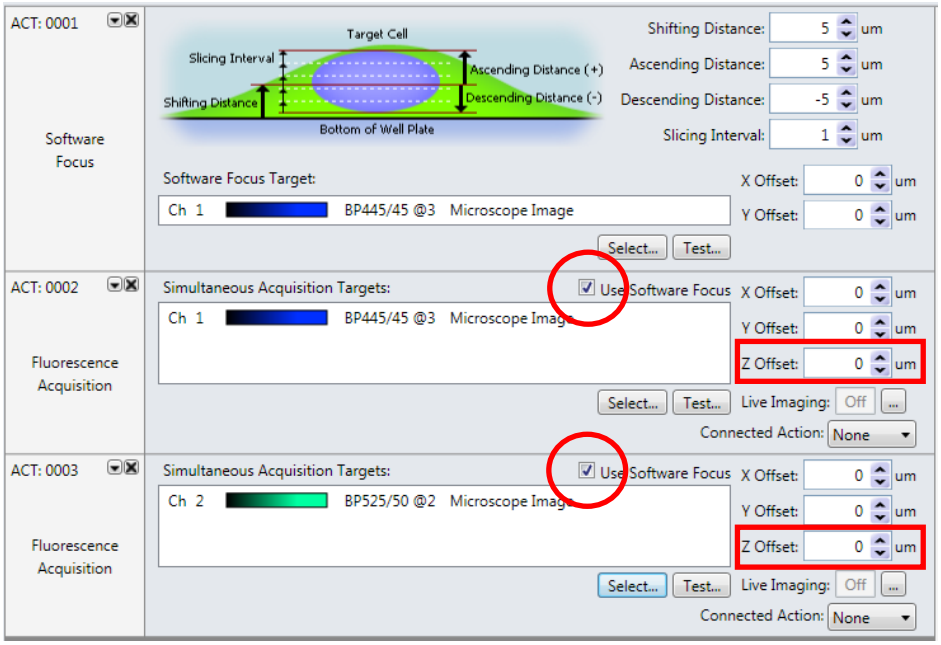
Confocal imaging is performed on the software focus plane at two wavelengths (405 nm, 488 nm).

1) Open the measurement setting file edit screen. (Refer to 5.2)
2) Enter the application name. (Refer to 5.4)

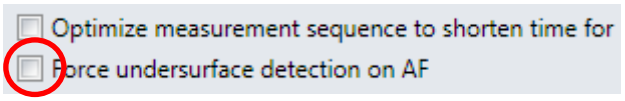
3) Set the imaging channels. (Refer to 5.5)
   Ch1 Method : Confocal Fluorescence 405/488/640nm
   Acquisition : BP445/45 Light Source : 405nm (30%)
   Ch2 Method : Confocal Fluorescence 405/488/640nm
   Acquisition : BP525/50 Light Source : 488nm (30%)



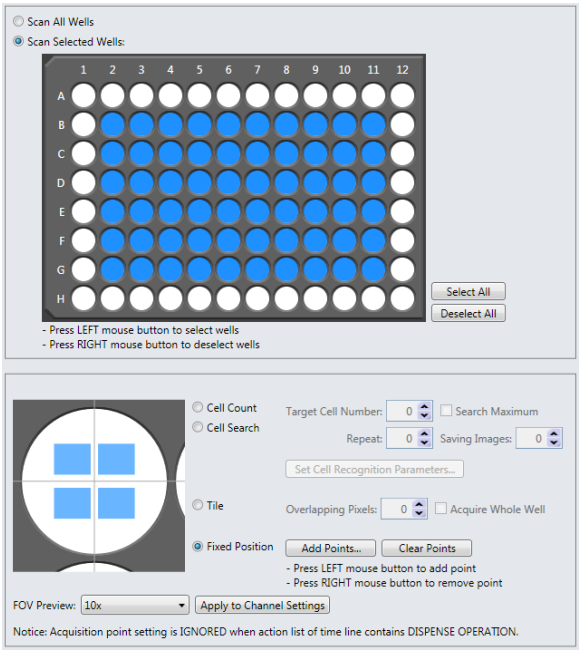
4) Set the items on the Action List tab. (Refer to 5.7)
   Software focus is applied to Ch1. Images are captured for Ch1 and Ch2 on the software focus plane. To capture images on the software focus plane, set “0” under “Z Offset” for “Fluorescence Acquisition.”



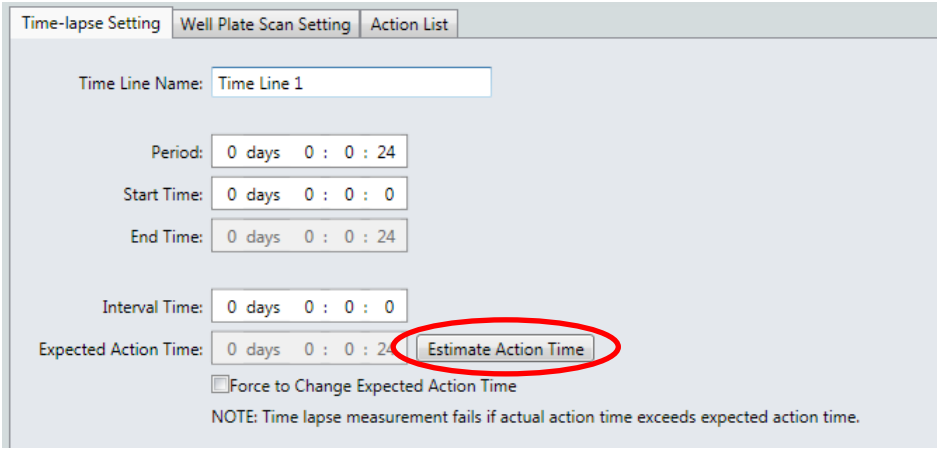
Set “Force undersurface detection on AF”. Normally, uncheck this box. If high-speed time-lapse imaging with operating autofocus is performed after software focus, check this box.



5) Set the imaging wells and view field on the Well Plate Scan Setting tab. (Refer to 5.6)



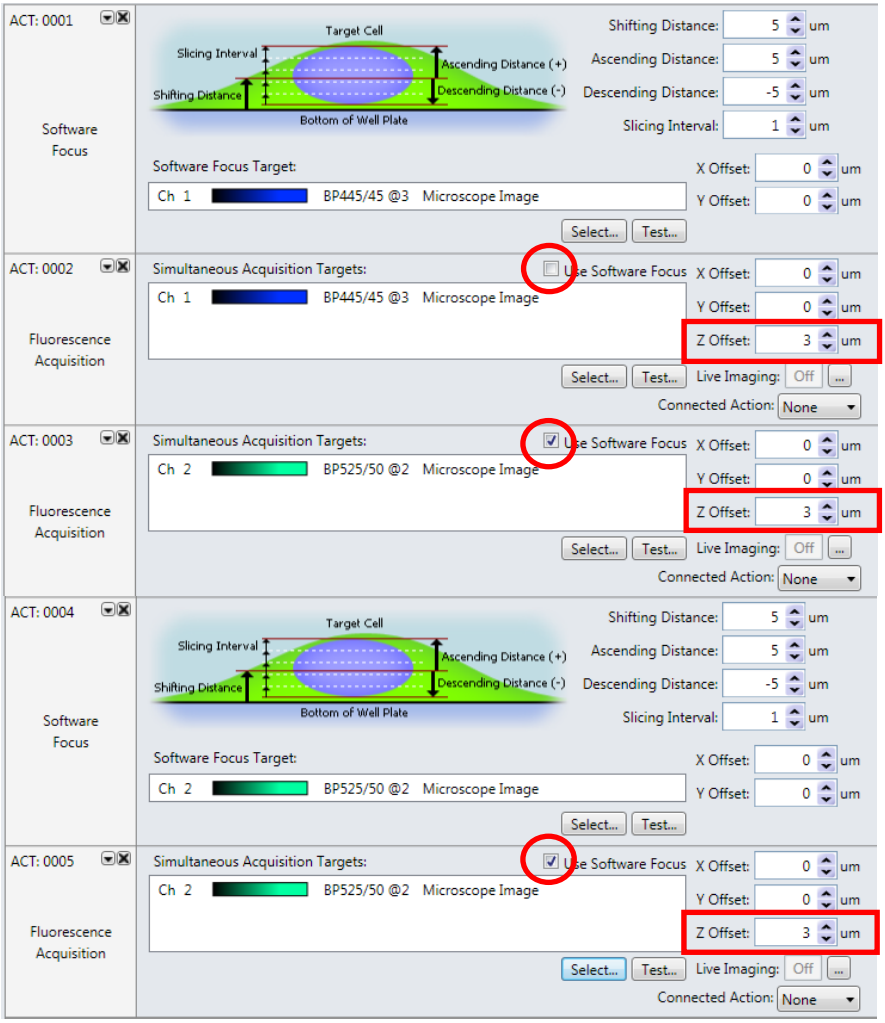
6) Click “Estimate Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3)



7) Save the measurement setting file. (Refer to 5.11)

MEMO

● Images are acquired based on the last software focus setting if multiple software focus settings are set.



ACT0001 : Get the software-focused position scanned in the range by Ch1.
ACT0002 : Acquire the Ch1 image at the Z position where 3um are added to the auto-focused position.
ACT0003 : Acquire the Ch2 image at the Z position where 3um are added to the software-focused position by Ch1.
ACT0004 : Get the software-focused position scanned in the range by Ch2.
ACT0005 : Acquire the Ch2 image at the Z position where 3um are added to the software-focused position by Ch2.

3D Imaging from the Software Focus Plane

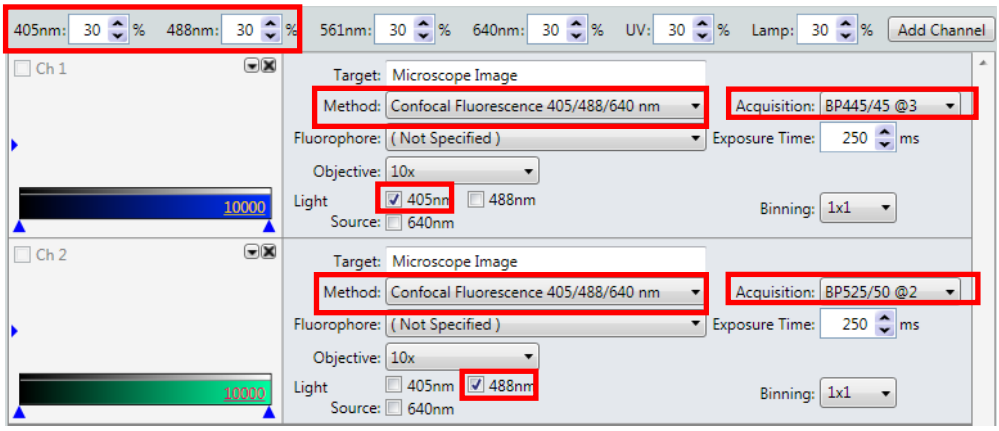
3D imaging is performed at two wavelengths (405 nm, 488 nm) by using the software focus plane as a reference.

1) Open the measurement setting file edit screen. (Refer to 5.2)

2) Enter the application name. (Refer to 5.4)

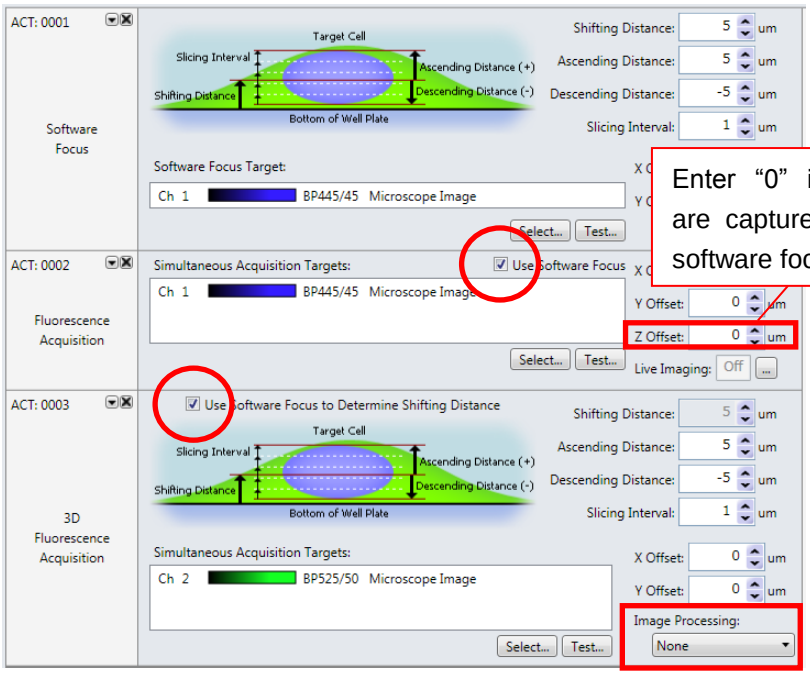
3) Set the imaging channels. (Refer to 5.5)

Ch1 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP445/45 Light Source : 405nm (30%)
Ch2 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP525/50 Light Source : 488nm (30%)



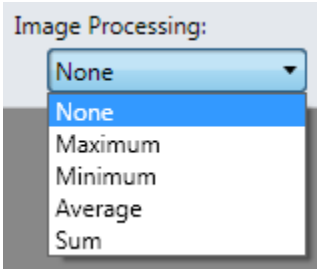
4) Set the items on the Action List tab. (Refer to 5.7)

Software focus is applied to Ch1. Images are captured on the software focus plane for Ch1, and 3D imaging is performed for Ch2. Select the “Use Software Focus to Determine Shifting Distance” check box.

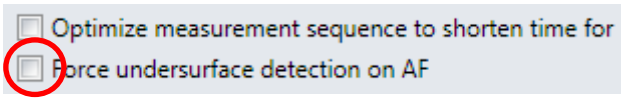


Enter “0” if images are captured on the software focus plane.

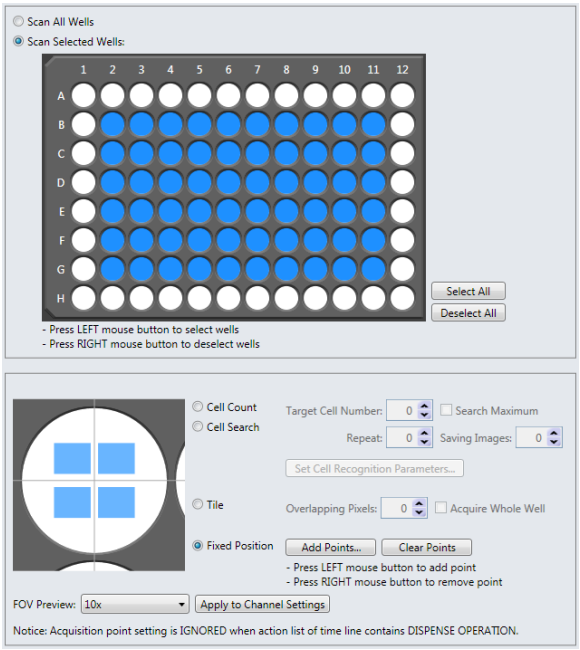
Select an output method for Z images from “Image Processing.”
(Refer to 5.1 and 6.2.)



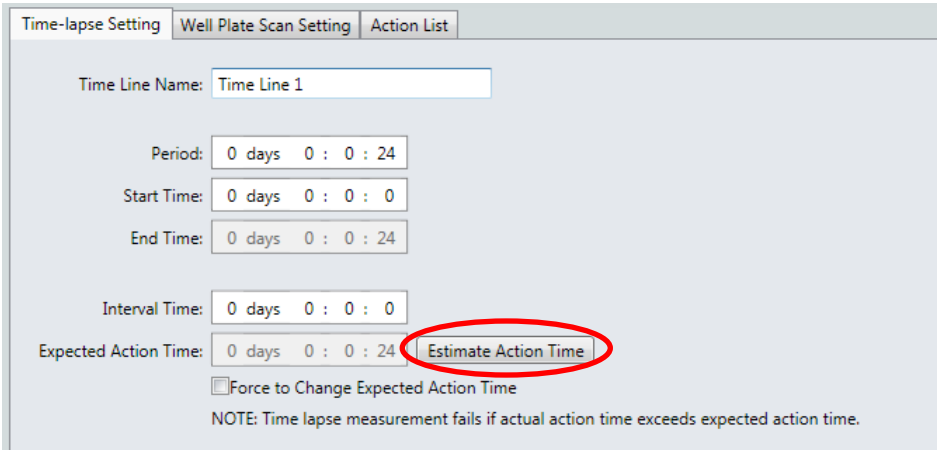
Uncheck “Force undersurface detection on AF”.



5) Set the imaging wells and view field on the Well Plate Scan Setting tab.
(Refer to 5.6)



6) Click “Estimate Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3)



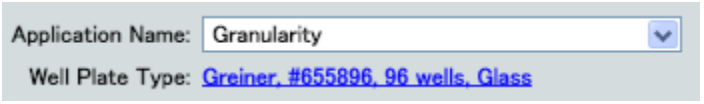
7) Save the measurement setting file. (Refer to 5.11)

7.3. Confocal Imaging and Bright-field Imaging of the Same View Field

Confocal imaging is performed at two wavelengths (405 nm, 488 nm) and then imaging of the same view field is performed in the bright-field mode.

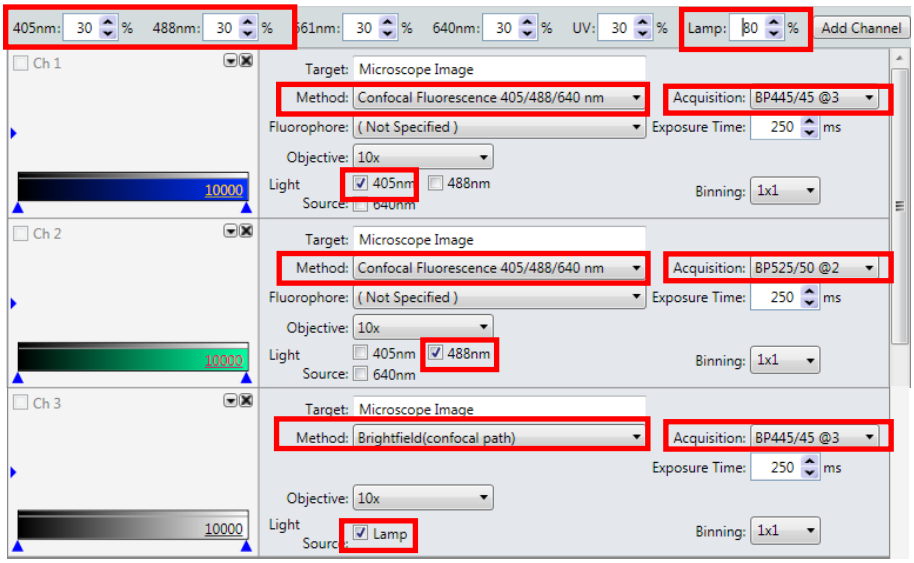
1) Open the measurement setting file edit screen. (Refer to 5.2)

2) Enter the application name. (Refer to 5.4)

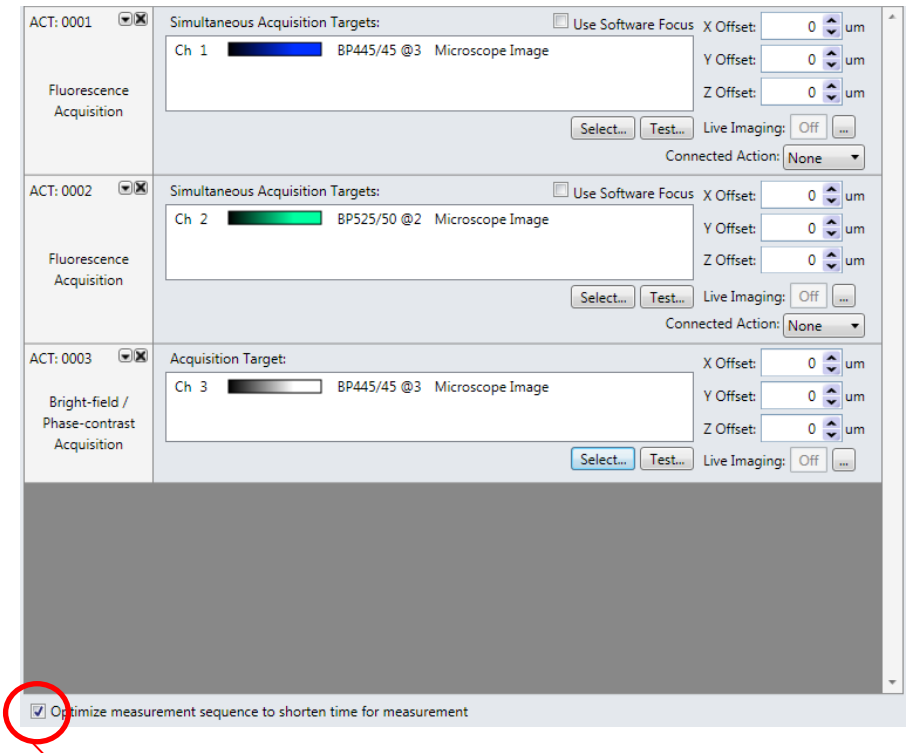


3) Set the imaging channels. (Refer to 5.5)

Ch1 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP445/45 Light Source : 405nm (30%)
Ch2 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP525/50 Light Source : 488nm (30%)
Ch3 Method : Brightfield(Confocal path)
Acquisition : BP525/50 Light Source : Lamp (80%)



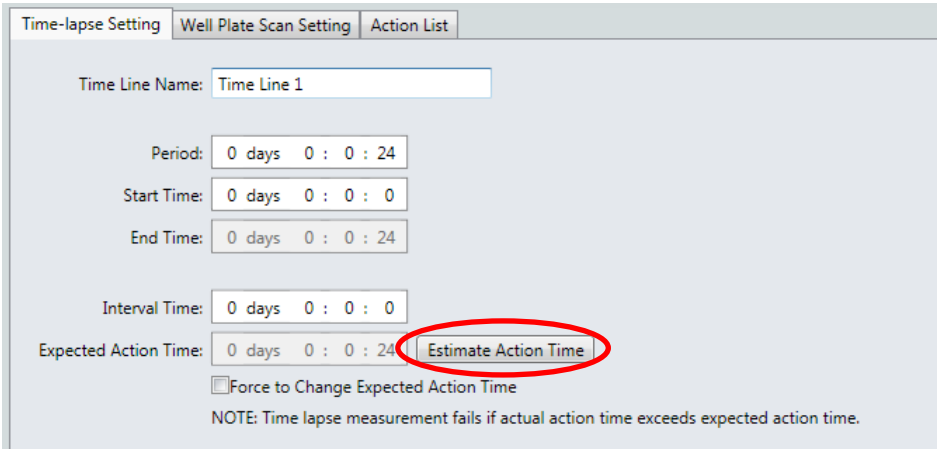
4) Set the items on the Action List tab. (Refer to 5.7)
Set Ch1 and Ch2 for the Fluorescence Acquisition tasks, respectively.
Set Ch3 for the Bright-field/Phase-contrast Acquisition task.



Check here to acquire hole well plate by the confocal fluorescence and then by bright field.
If the checkbox is unchecked, each image data is acquired at the same timing in the case that there are the differences of optical method and objective lens settings in the action list with switching optics..

5) Set the imaging wells and view field on the Well Plate Scan Setting tab. (Refer to 5.6)

6) Click “Estimate Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3)



7) Save the measurement setting file. (Refer to 5.11)

7.4. Imaging by Simultaneous Laser Emissions at Multi Wavelengths

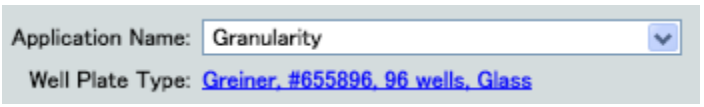
Images are acquired by emitting laser beams of multi wavelengths simultaneously. (Multi-camera model only)

Confocal Imaging by Simultaneous Laser Emissions at 3 Wavelengths

Confocal imaging is performed by emitting laser beams of three wavelengths (405 nm, 488 nm and 640 nm) simultaneously.

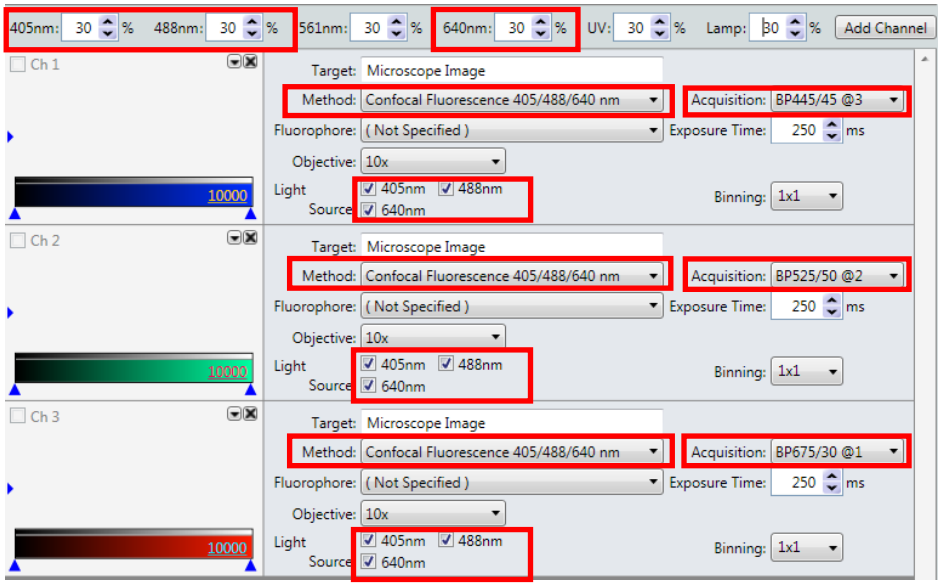
1) Open the measurement setting file edit screen. (Refer to 5.2)

2) Enter the application name. (Refer to 5.4)



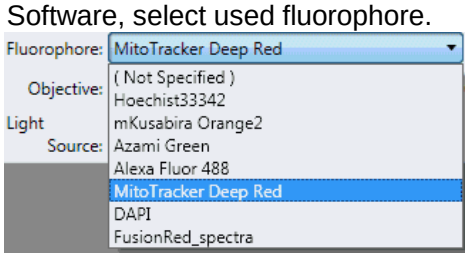
3) Set the imaging channels. (Refer to 5.5)

Ch1 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP445/45 Light Source : 405nm, 488nm, 640nm (30%)
Ch2 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP525/50 Light Source : 405nm, 488nm, 640nm (30%)
Ch3 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP675/30 Light Source : 405nm, 488nm, 640nm (30%)

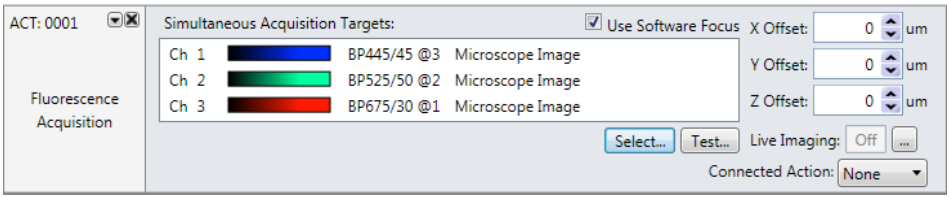


MEMO

● If performing crosstalk correction by Image Correction Software, select used fluorophore.

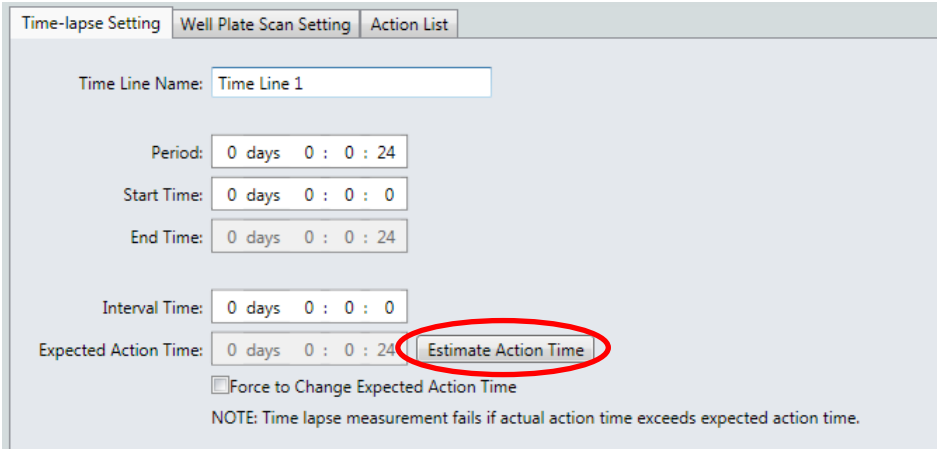


4) Set the items on the Action List tab. (Refer to 5.7)
Specify three wavelengths for one Fluorescence Acquisition task.



5) Set the imaging wells and view field on the Well Plate Scan Setting tab. (Refer to 5.6)

6) Click “Estimate Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3)



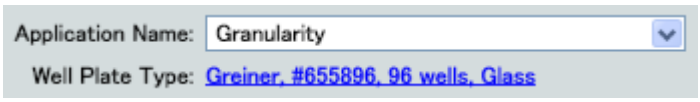
7) Save the measurement setting file. (Refer to 5.11)

3D Imaging by Simultaneous Laser Emissions at 3 Wavelengths

3D imaging is performed by emitting laser beams of three wavelengths (405 nm, 488 nm, 640 nm) simultaneously.

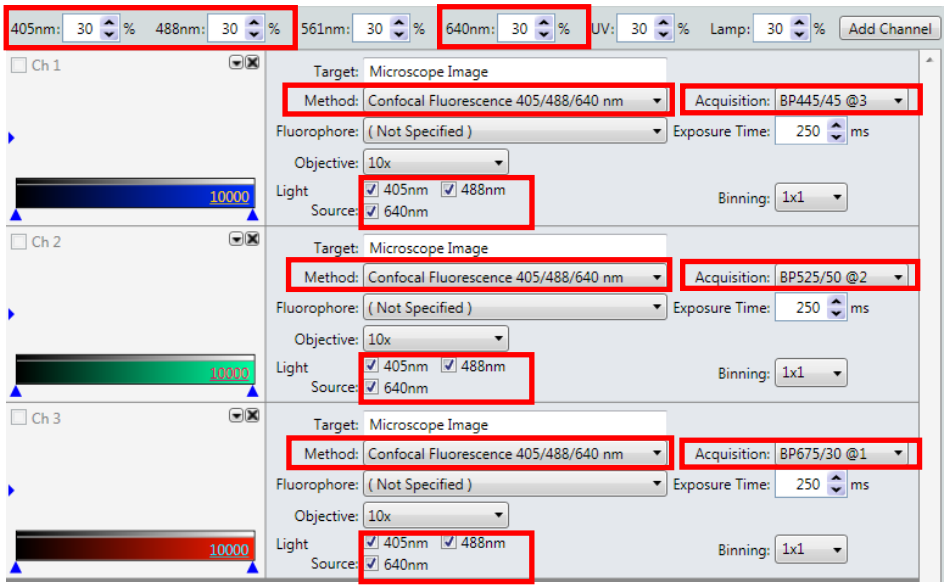
1) Open the measurement setting file edit screen. (Refer to 5.2)

2) Enter the application name. (Refer to 5.4)



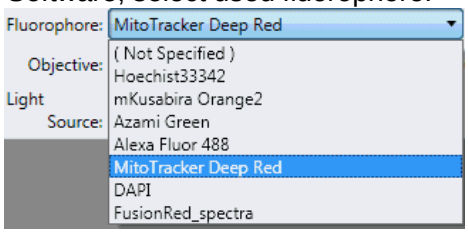
3) Set the imaging channels. (Refer to 5.5)

Ch1 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP445/45 Light Source : 405nm、488nm、640nm (30%)
Ch2 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP525/50 Light Source : 405nm、488nm、640nm (30%)
Ch3 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP675/30 Light Source : 405nm、488nm、640nm (30%)

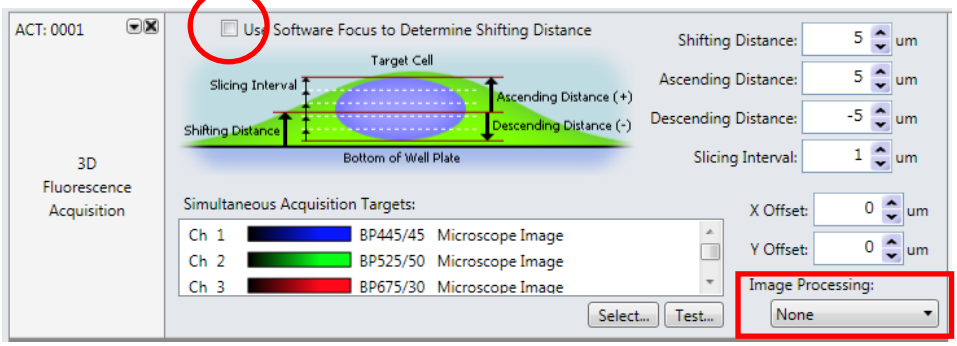


MEMO

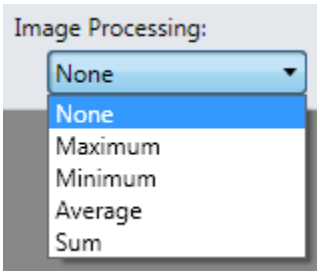
● If performing crosstalk correction by Image Correction Software, select used fluorophore.



4) Set the items on the Action List tab. (Refer to 5.7)
Specify three wavelengths for one 3D Fluorescence Acquisition task.
Unselect the “Use Software Focus to Determine Shifting Distance” check box.

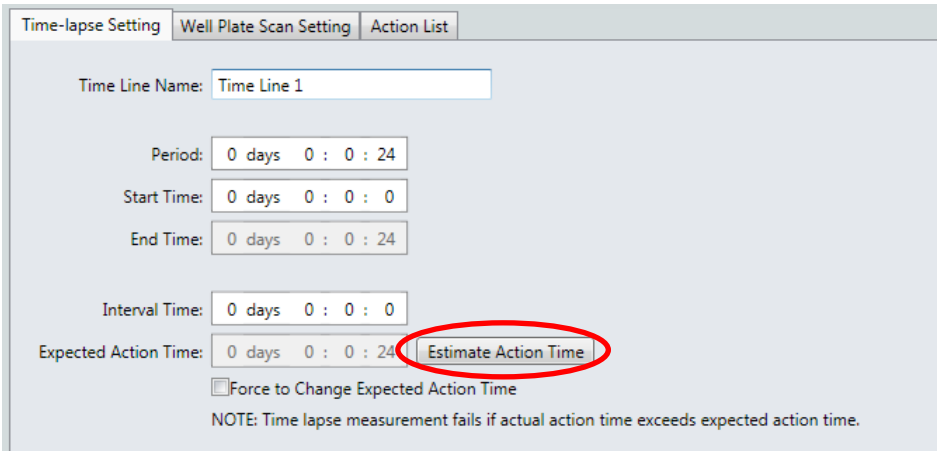


Select an output method for Z images from “Image Processing.”
(Refer to 5.1 and 6.2.)



5) Set the imaging wells and view field on the Well Plate Scan Setting tab. (Refer to 5.6)

6) Click “Estimate Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3.)



7) Save the measurement setting file. (Refer to 5.11)

7.5. High-speed Time-lapse Imaging per Well

In Case without Performing Autofocus during Imaging

MEMO

● In this case, perform autofocus before the first image acquisition. Focus may shift gradually by thermal expansion of machine.

High-speed time-lapse imaging is performed with a laser beam of 488 nm in wavelength at an imaging interval of 400 ms and imaging time of 30 seconds per well, with dispensing performed five seconds after the start of imaging.

1) Open the measurement setting file edit screen. (Refer to 5.2)

2) Enter the application name. (Refer to 5.4)

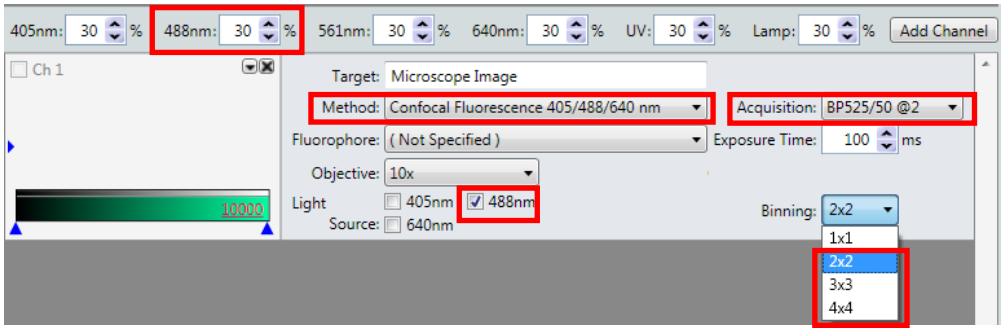
3) Set the imaging channels. (Refer to 5.5)

Ch1 Method：Confocal Fluorescence 405/488/640nm

Acquisition：BP525/50 Light Source：488nm (30%) Binning: 2

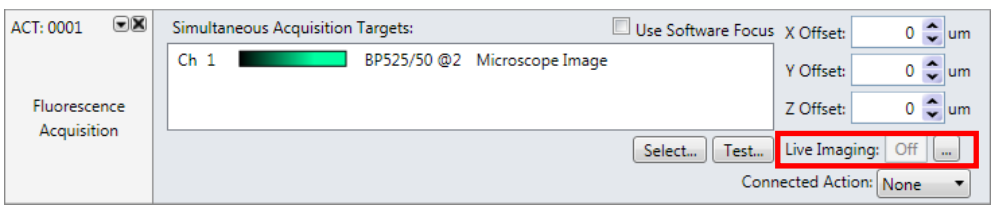
MEMO

●Since a very large amount of image data will be acquired in high-speed time-lapse imaging, the binning setting more than “2x2” is recommended.

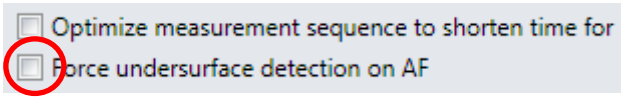


4) Set the items on the Action List tab. (Refer to 5.7)

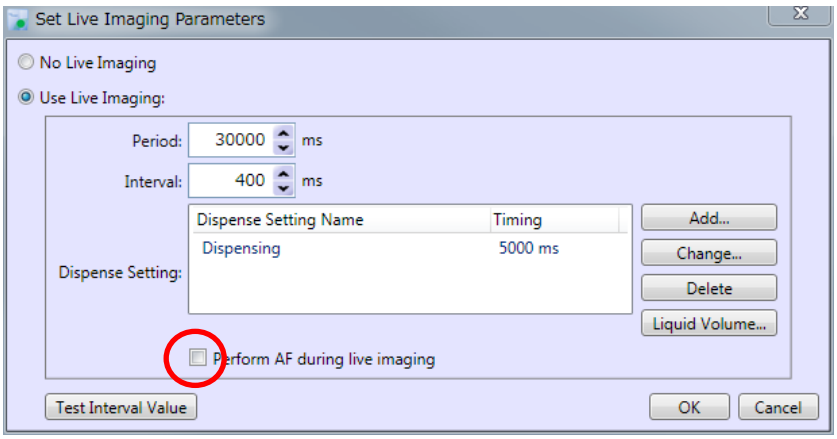
To perform fluorescence imaging, set “Fluorescence Acquisition” task. To perform bright field/phase contrast imaging, set a “Bright-field/ Phase-contrast Acquisition” task.



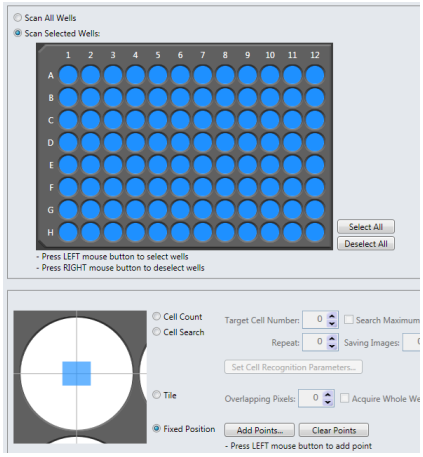
Uncheck “Force undersurface detection on AF”.



5) Set high-speed time-lapse imaging. Uncheck “Performing AF during live imaging”. (Refer to 5.8)



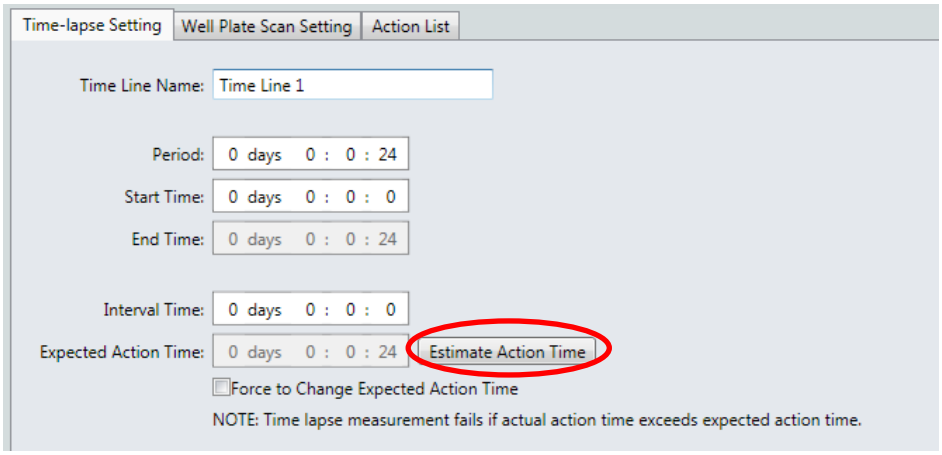
6) Set the imaging wells and view field on the Well Plate Scan Setting tab. (Refer to 5.6)



MEMO

- When images are captured by high-speed time-lapse imaging, the view field covers only one point at the center of the well.
- Select same acquisition points to correspond to the well-plate map specified by dispensing setting file (refer to 5.12) for Assay plate.

7) Click “Estimate Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3.)



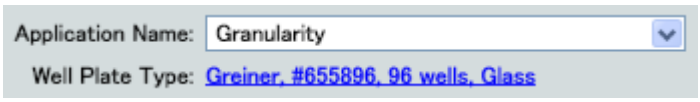
8) Save the measurement setting file. (Refer to 5.11)

In Case with Performing Autofocus during Imaging

High-speed time-lapse imaging is performed with a laser beam of 488 nm in wavelength at an imaging interval of 10 seconds and imaging time of 5 minutes per well, with dispensing performed 20 seconds after the start of imaging.

1) Open the measurement setting file edit screen. (Refer to 5.2)

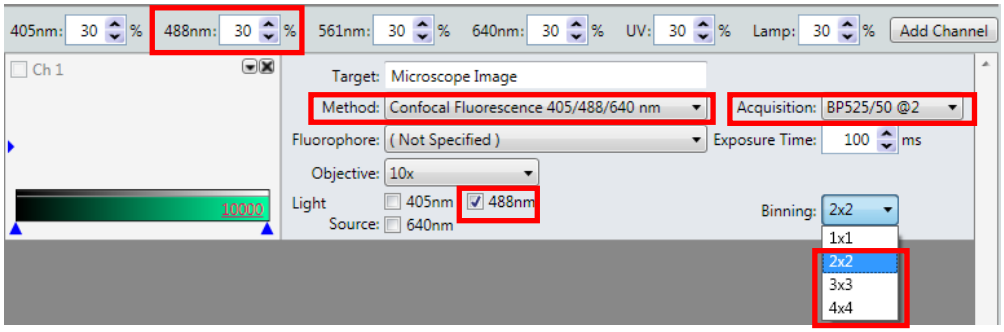
2) Enter the application name. (Refer to 5.4)



3) Set the imaging channels. (Refer to 5.5)
Ch1 Method：Confocal Fluorescence 405/488/640nm
Acquisition：BP525/50 Light Source：488nm (30%) Binning: 2

MEMO

●Since a very large amount of image data will be acquired in high-speed time-lapse imaging, the binning setting more than “2x2” is recommended.

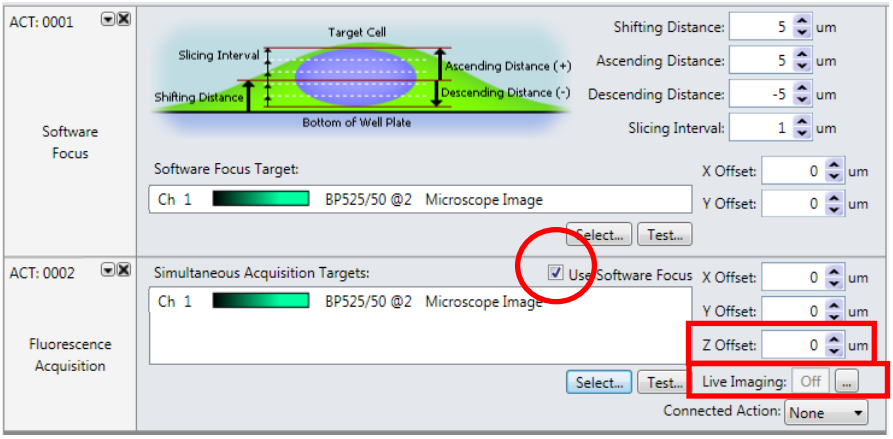


4) Set the items on the Action List tab. (Refer to 5.7)

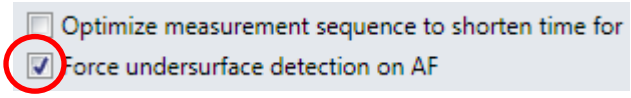
To perform fluorescence imaging, set “Fluorescence Acquisition” task. To perform bright field/phase contrast imaging, set a “Bright-field/ Phase-contrast Acquisition” task.

Software focus is applied to Ch1. Images are captured for Ch1 on the software focus plane. Check “Use Software Focus” and set “0” under “Z Offset” for “Fluorescence Acquisition.”

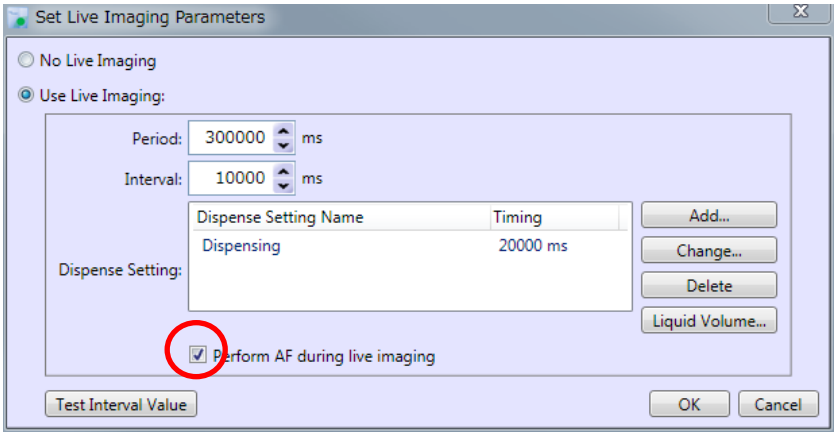
Click “Live Imaging”



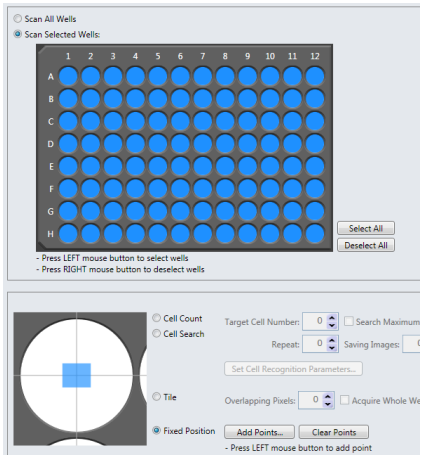
Check “Force undersurface detection on AF”.



5) Set high-speed time-lapse imaging. Check “Performing AF during live imaging”. (Refer to 5.8)



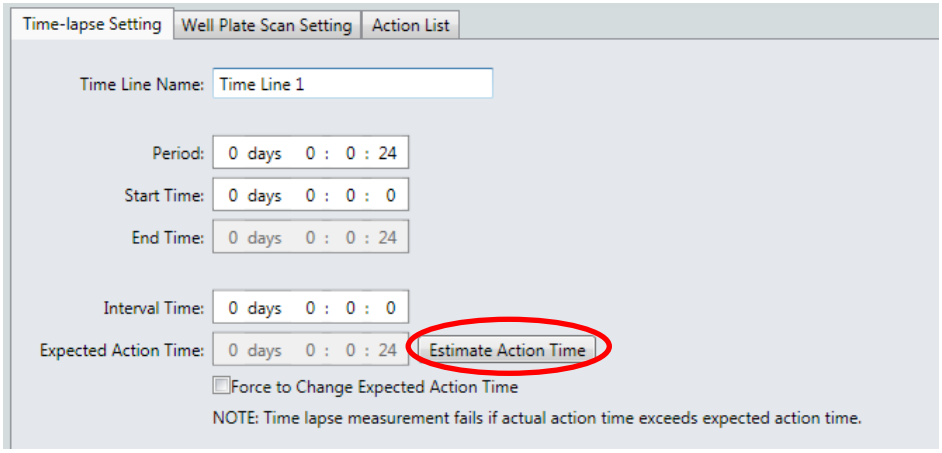
6) Set the imaging wells and view field on the Well Plate Scan Setting tab. (Refer to 5.6)



MEMO

- When images are captured by high-speed time-lapse imaging, the view field covers only one point at the center of the well.
- Select same acquisition points to correspond to the well-plate map specified by dispensing setting file (refer to 5.12) for Assay plate.

7) Click “Estimate Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3.)



8) Save the measurement setting file. (Refer to 5.11)

7.6. Time-lapse Imaging per Plate without Dispensing

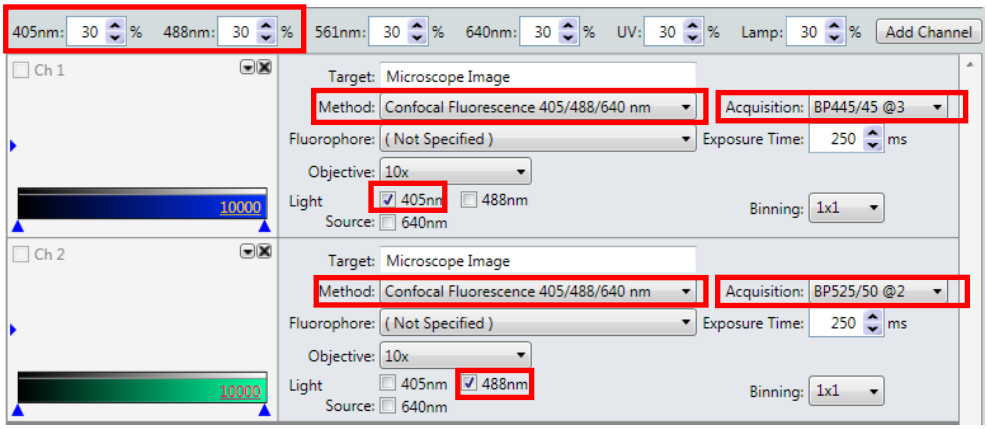
Time-lapse imaging is performed for one assay plate at 15-minute intervals over a period of 50 minutes.

1) Open the measurement setting file edit screen. (Refer to 5.2)

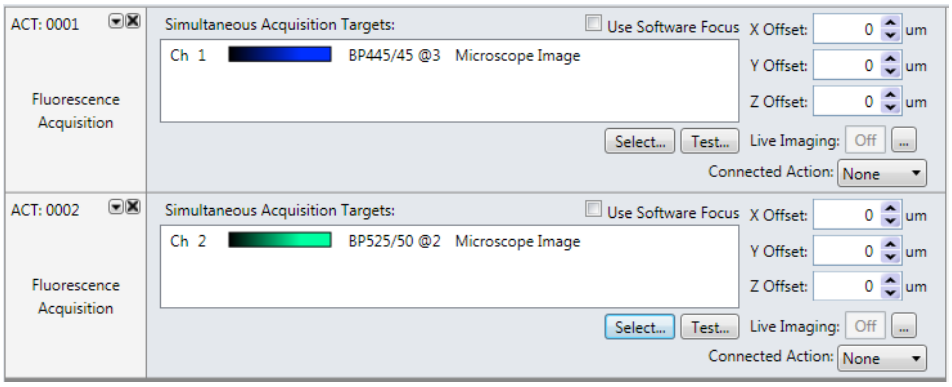
2) Enter the application name. (Refer to 5.4)

3) Set the imaging channels. (Refer to 5.5)

Ch1 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP445/45 Light Source : 405nm (30%)
Ch2 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP525/50 Light Source : 488nm (30%)

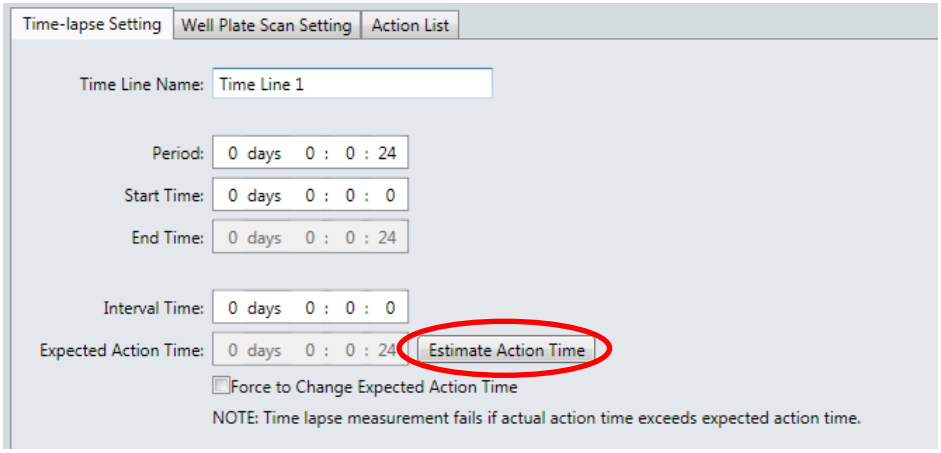


4) Set the items on the Action List tab. (Refer to 5.7)

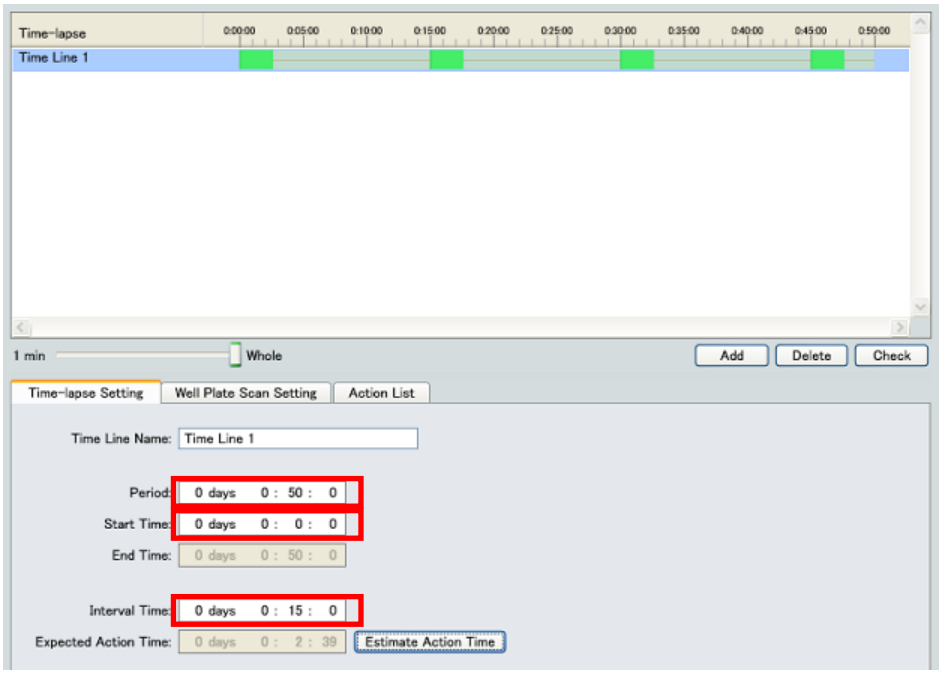


5) Set the imaging wells and view field on the Well Plate Scan Setting tab. (Refer to 5.6.)

6) Click “Estimate Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3)



7) Enter the values of “Interval Time,” “Start Time” and “Period.” (Refer to 5.3) (Set the interval time to 15 minutes, the start time to 0 minute and the period to 50 minutes.)



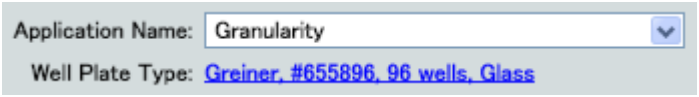
8) Save the measurement setting file. (Refer to 5.11)

7.7. Time-lapse Imaging per Plate with Dispensing

One assay plate is measured once, and 15 minutes thereafter the dispensing is performed from one source plate. After 40 minutes, time-lapse imaging is performed for the assay plate at 15-minute intervals over a period of 50 minutes.

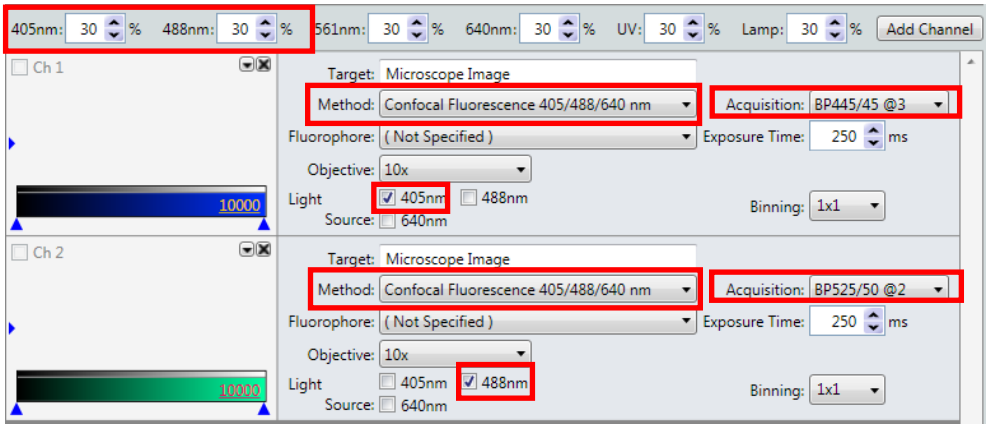
1) Open the measurement setting file edit screen. (Refer to 5.2)

2) Enter the application name. (Refer to 5.4)

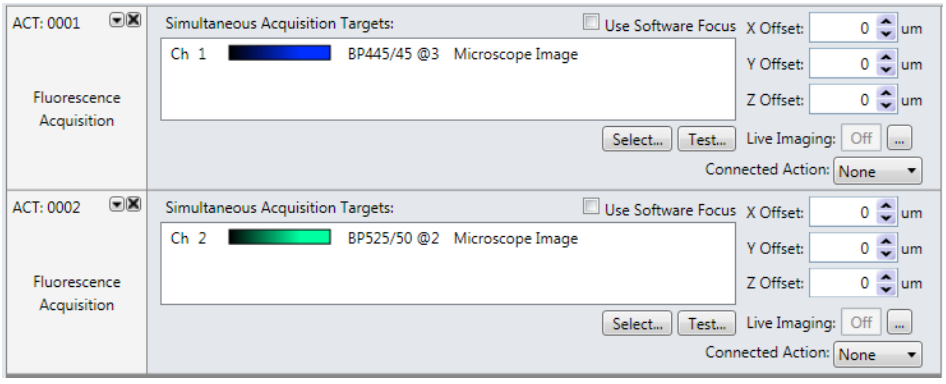


3) Set the imaging channels. (Refer to 5.5)

Ch1 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP445/45 Light Source : 405nm (30%)
Ch2 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP525/50 Light Source : 488nm (30%)

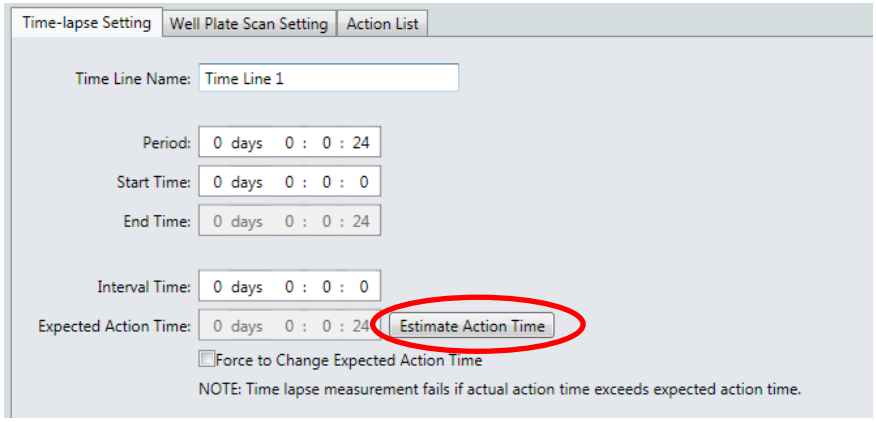


4) Set the items on the Action List tab. (Refer to 5.7)

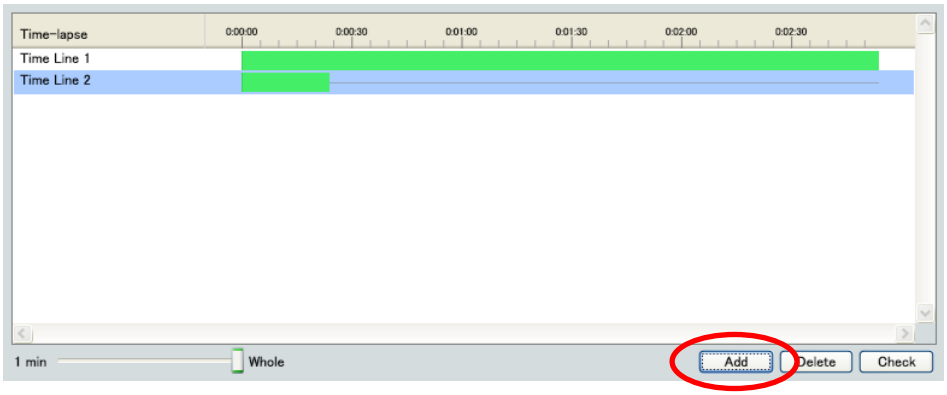


5) Set the imaging wells and view field on the well Plate Scan Setting tab. (Refer to 5.6.)

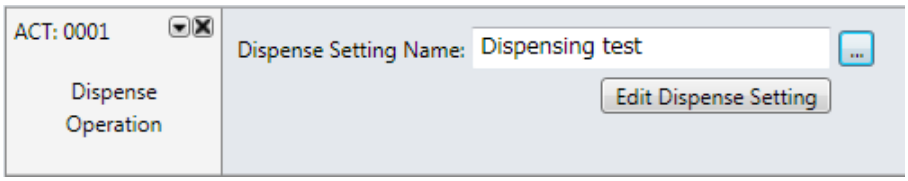
6) Click “Estimate Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3)



7) Create a dispensing time line. Click “Add” and create a new time line. (Refer to 5.3)



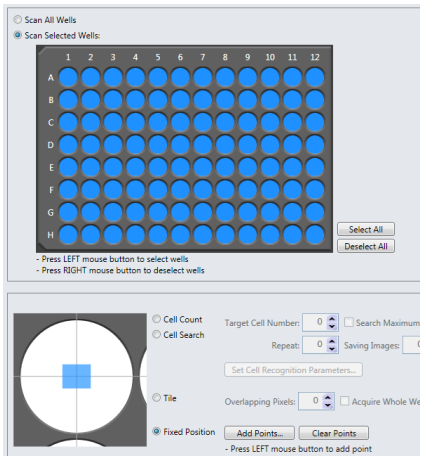
8) Set the items on the Action List tab. (Refer to 5.7)
Refer to 7.12 for example of dispensing setting



MEMO

- In case of setting “Dispense Operation”, assign only “Dispense Operation” in Action List. If other actions like “Fluorescence Acquisition” should be set, assign in other timeline.

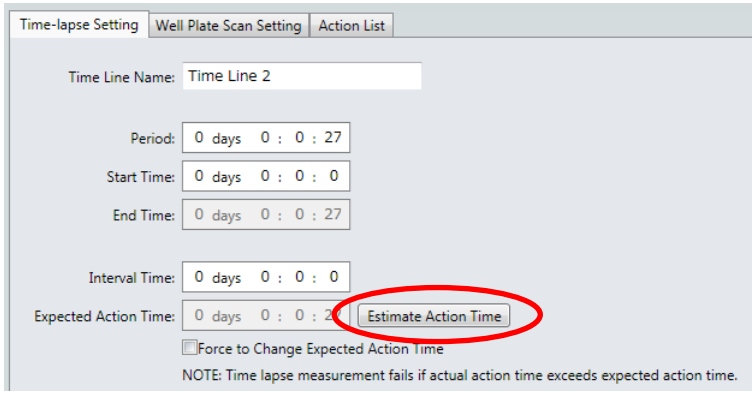
9) Set the wells to dispense. (Refer to 5.6)
Solution is dispensed once for each well. The Acquisition Points settings are not reflected.



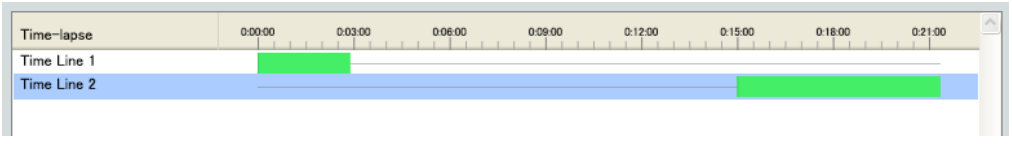
MEMO

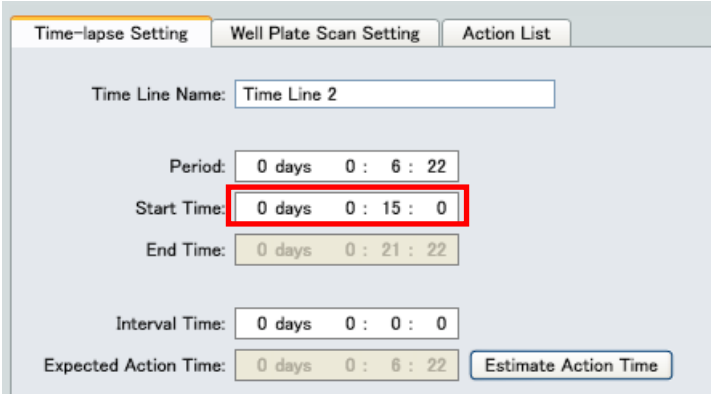
●Select same acquisition points to correspond to the well-plate map specified by dispensing setting file (refer to 5.12) for Assay plate.

10) Click “Estimate Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3)

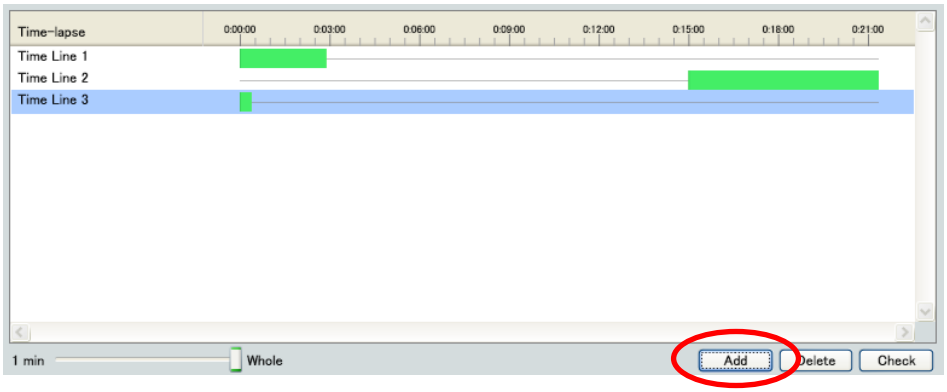


11) Enter the value of “Start Time” for Time Line 2. (Refer to 5.3)

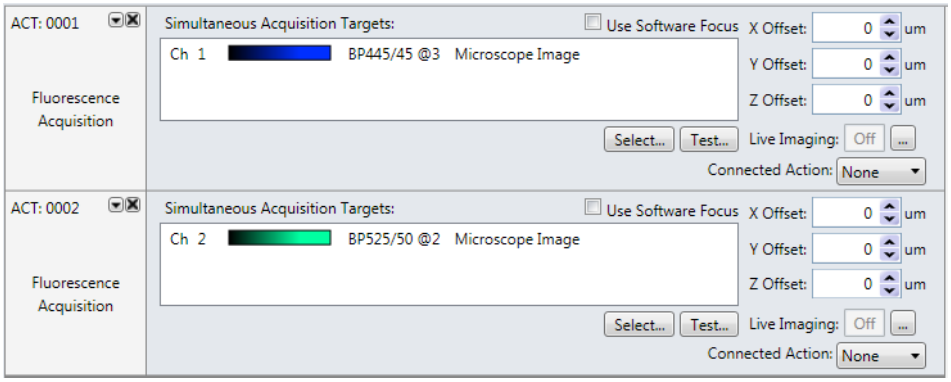




12) Create a new time-lapse time line. Click "Add" and create a new time line. (Refer to 5.3)

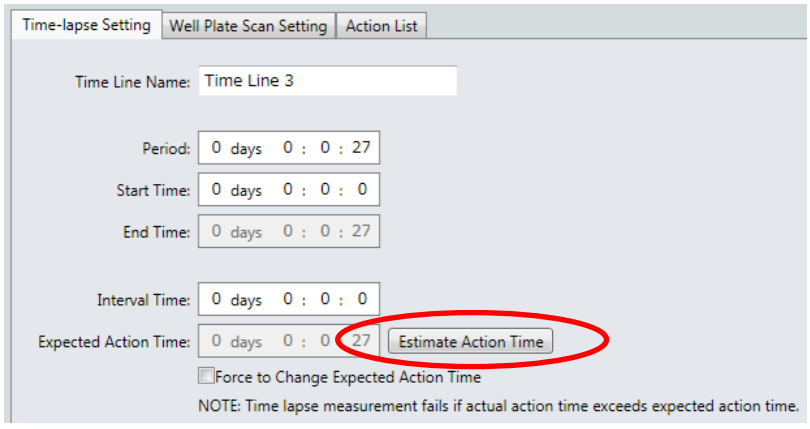


13) Set the items on the Action List tab. (Refer to 5.7)

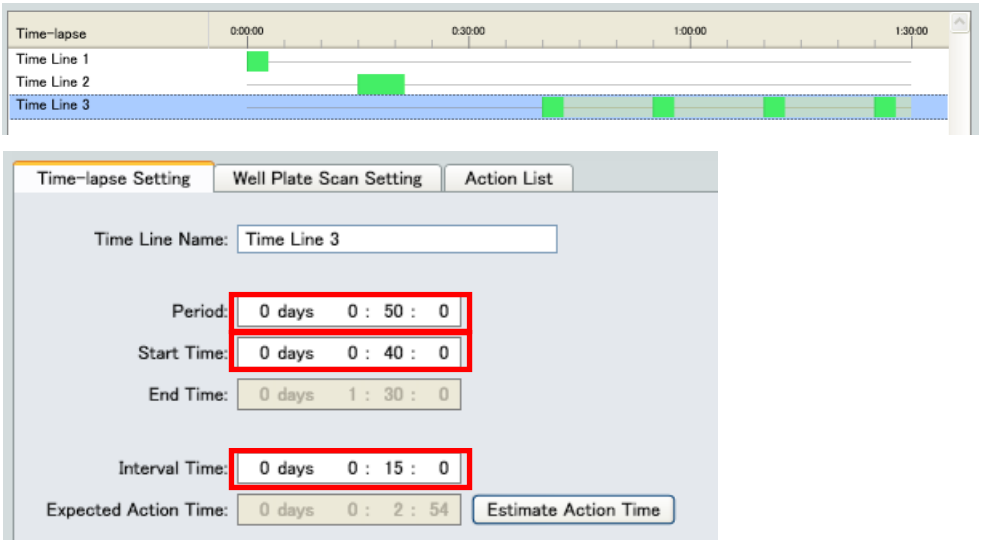


14) Set the imaging wells and view field on the Well Plate Scan Setting tab. (Refer to 5.6)

15) Click “Estimate Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3)



16) Enter the values of “Start Time” “Interval Time” and “Period” for Time Line 3. (Refer to 5.3)



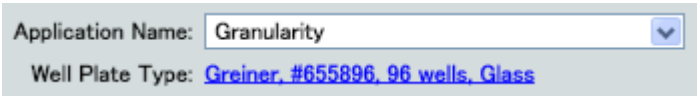
17) Save the measurement setting file. (Refer to 5.11)

7.8. Capturing Images According to the View Field Observed in the Test Preview

Capture images of the wells observed in the test preview according to the corresponding view field.

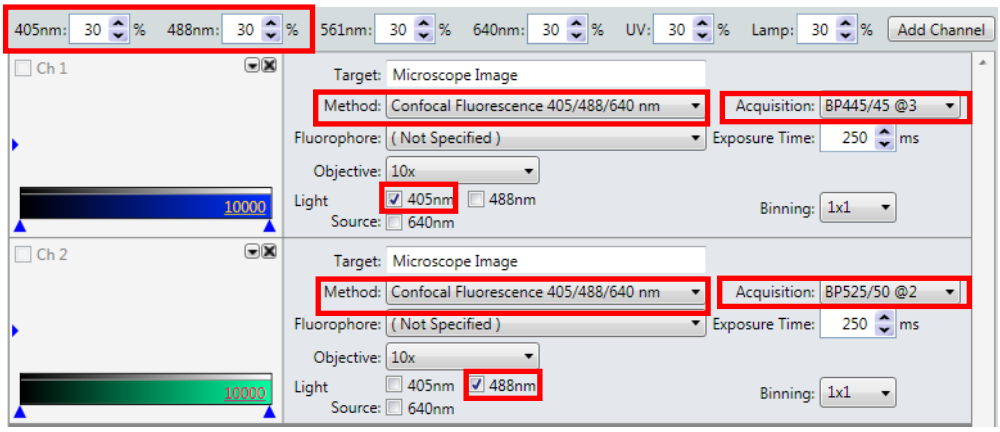
1) Open the measurement setting file edit screen. (Refer to 5.2)

2) Enter the application name. (Refer to 5.4)

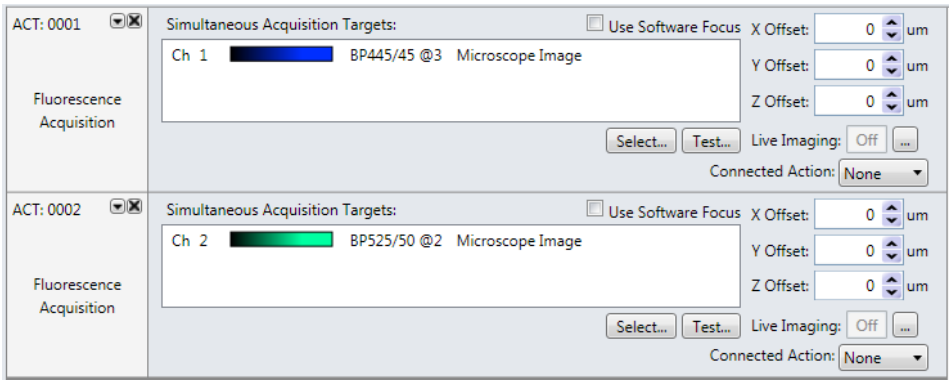


3) Set the imaging channels. (Refer to 5.5)

Ch1 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP445/45 Light Source : 405nm (30%)
Ch2 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP525/50 Light Source : 488nm (30%)

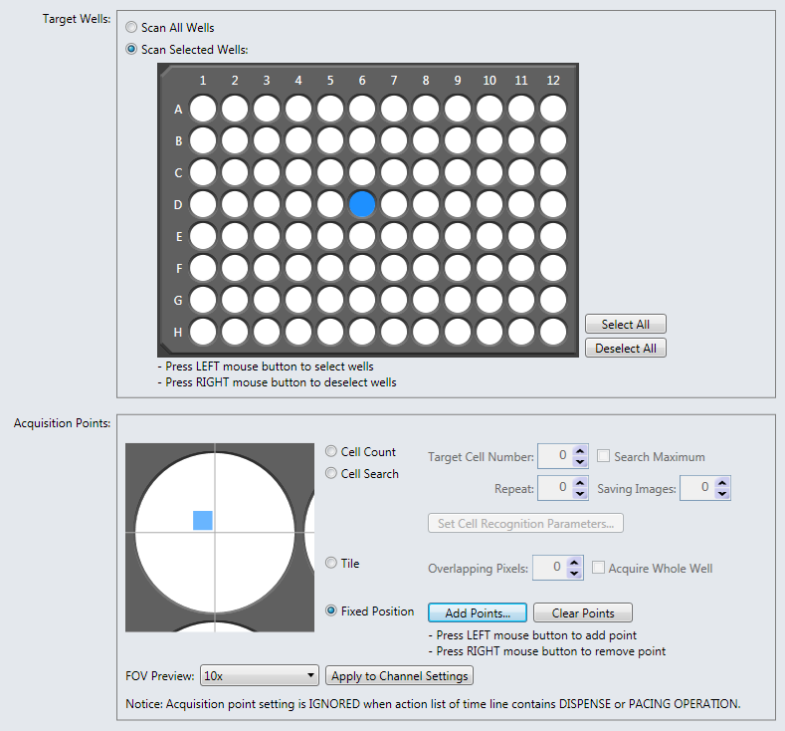


4) Set the items on the Action List tab. (Refer to 5.7)

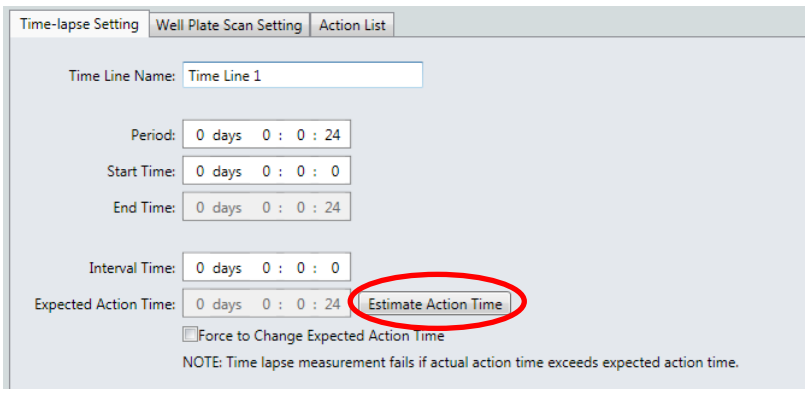


5) Display a test preview. (Refer to 5.9 and 6.2)
Before opening the test preview screen, select the “Synchronize Well Plate Scan Setting” check box. Specify the imaging wells and view field.

6) Confirm on the Well Plate Scan Setting tab that the wells and view field specified in the test preview are set. You can also add imaging wells and view fields. (Refer to 5.6)



7) Click “Estimate Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3)



8) Save the measurement setting file. (Refer to 5.11)

7.9. Imaging by Cell Count

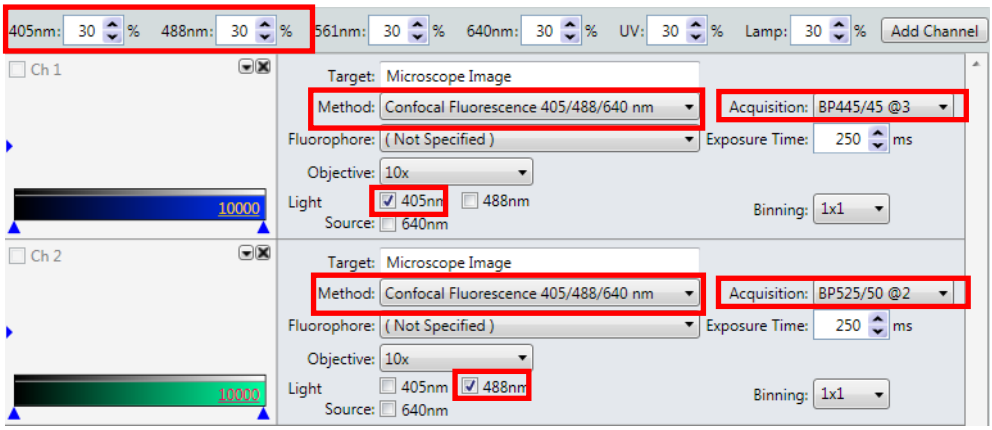
Images are captured repeatedly in the same well while moving through the acquisition points until the specified cell count is reached. Once the number of repetitions reaches the specified value, imaging is stopped and the system moves to the next well.

1) Open the measurement setting file edit screen. (Refer to 5.2)

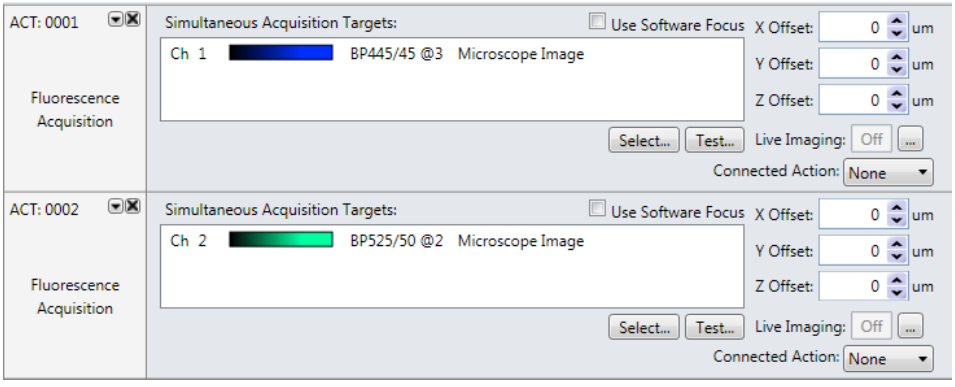
2) Enter the application name. (Refer to 5.4)

3) Set the imaging channels. (Refer to 5.5)

Ch1 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP445/45 Light Source : 405nm (30%)
Ch2 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP525/50 Light Source : 488nm (30%)

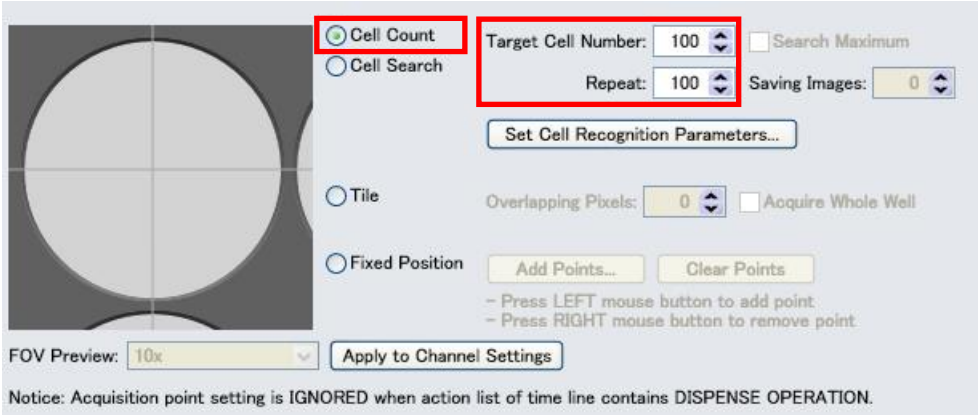


4) Set the items on the Action List tab. (Refer to 5.7)

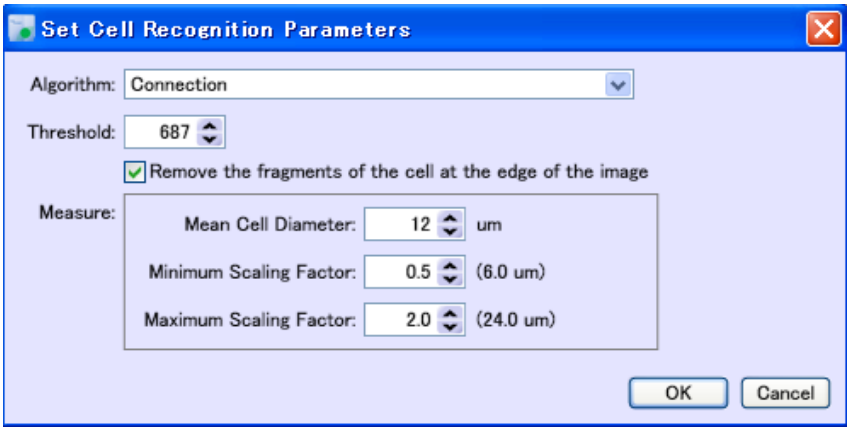


5) Set the imaging wells and view field on the Well Plate Scan Setting tab. (Refer to 5.6)

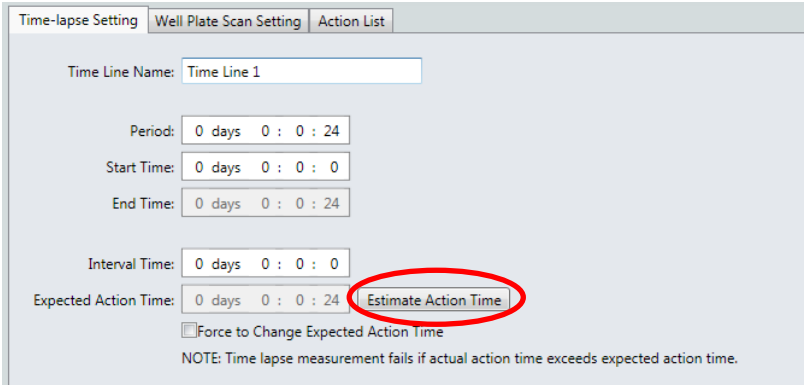
6) Select “Cell Count” on the Acquisition Points screen. (Refer to 5.6)



7) Set the cell recognition algorithm. (Refer to 5.10)



8) Click “Estimate Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3)



9) Save the measurement setting file. (Refer to 5.11)

7.10. Imaging by Water Immersion Lens

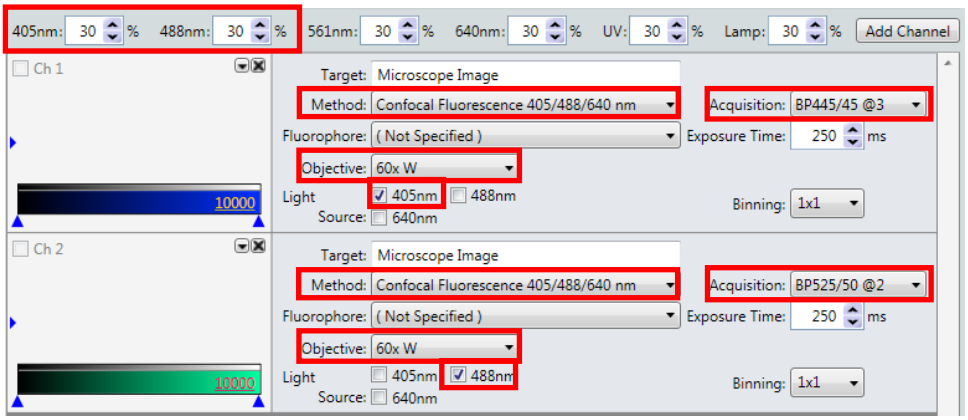
Confocal imaging is performed on the auto-focus plane at two wavelengths (405 nm, 488 nm) by water immersion lens.
(Water immersion lens model only)

1) Supply the water to the water immersion lenses. (Refer to 5.14)

2) Open the measurement setting file edit screen. (Refer to 5.2)

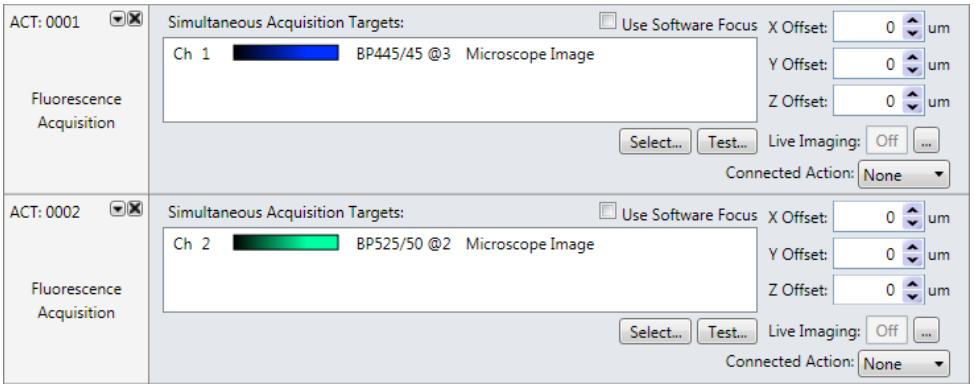
3) Enter the application name. (Refer to 5.4)

4) Set the imaging channels. (Refer to 5.5)
Ch1 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP445/45
Light Source : 405nm (30%)
Objective : 60x W
Ch2 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP525/50
Light Source : 488nm (30%)
Objective : 60x W



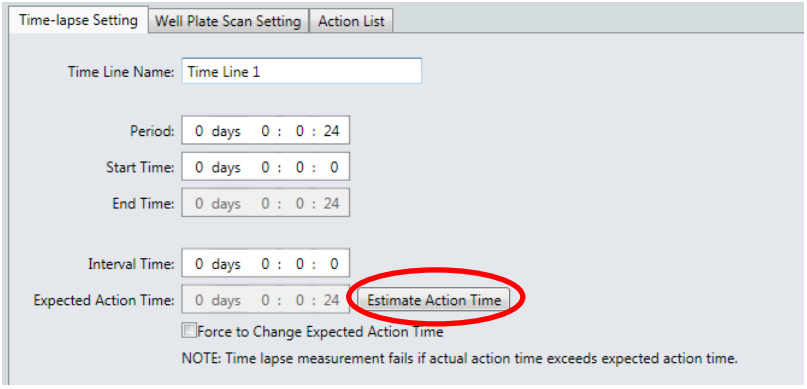
MEMO ●Select water immersion lenses in all imaging channels.

5) Set the items on the Action List tab. (Refer to 5.7)
Set Ch1 and Ch2 for the two Fluorescence Acquisition tasks, respectively.



6) Set the imaging wells and view field on the Well Plate Scan Setting tab. (Refer to 5.6)

7) Click “Estimate Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3)



8) Save the measurement setting file. (Refer to 5.11)

⚠ WARNING

- Wipe the water which attached to the bottom of the plate after the measurement by the water immersion lenses.

7.11. Imaging by 4-Laser Scanning (QUAD-DM and Dual filter model only)

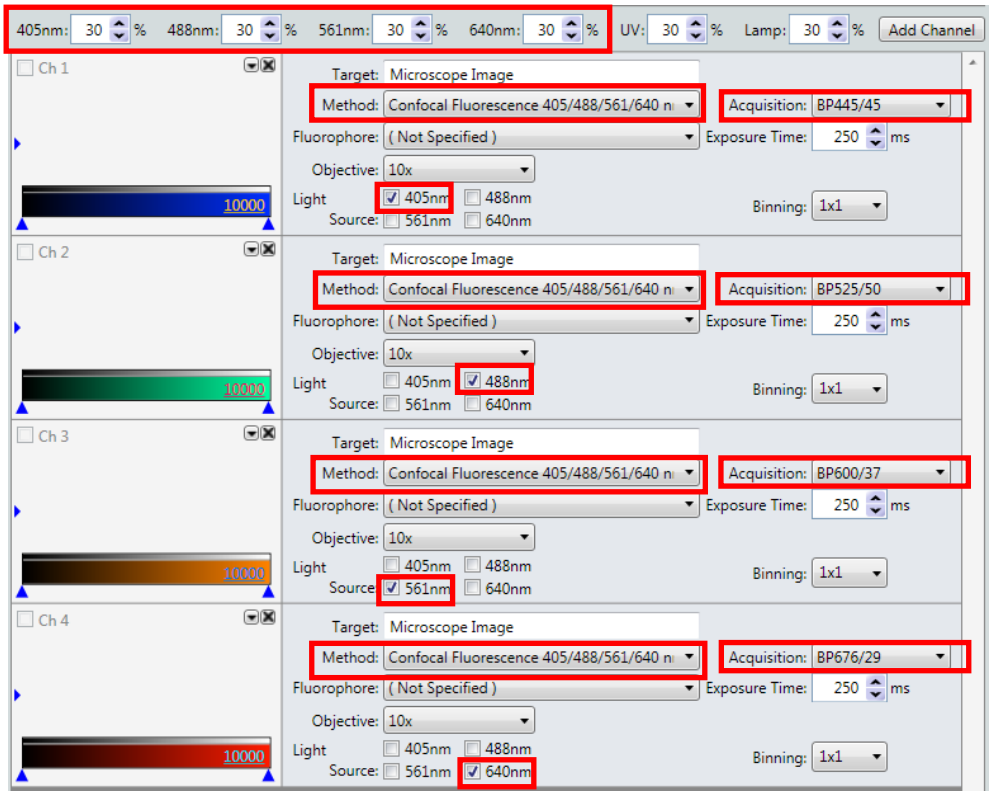
Confocal imaging is performed on the software focus plane at four wavelengths (405 nm, 488 nm, 561nm, 640nm).

1) Open the measurement setting file edit screen. (Refer to 5.2)

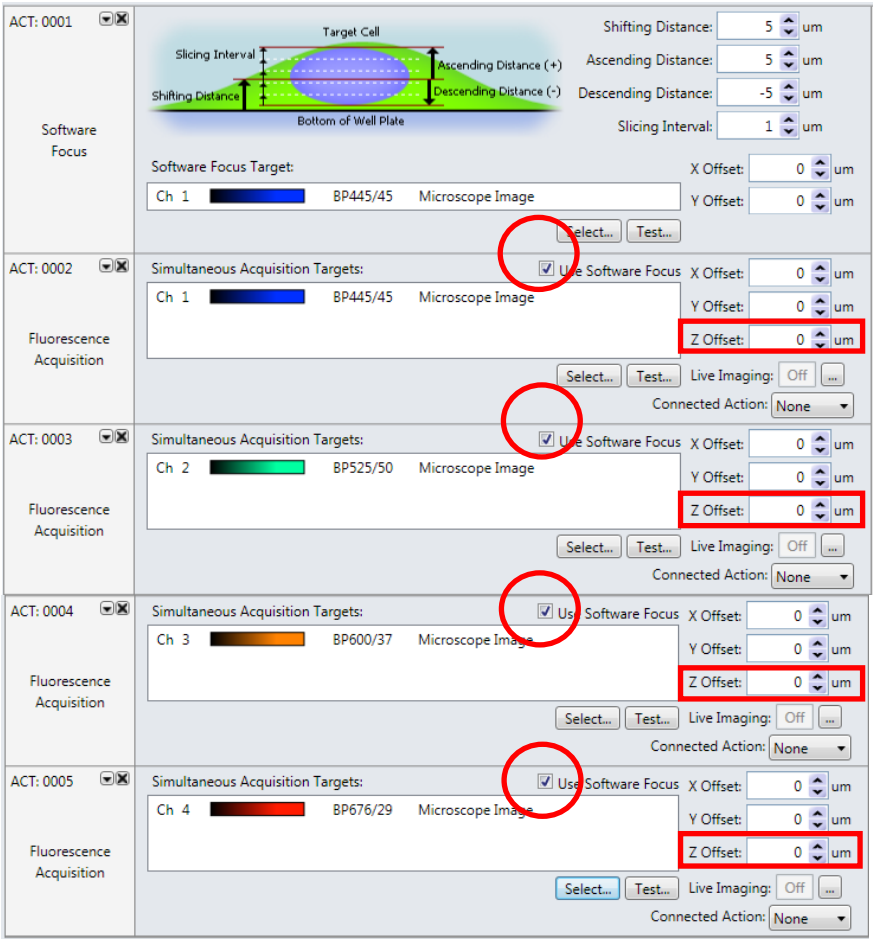
2) Enter the application name. (Refer to 5.4)

3) Set the imaging channels. (Refer to 5.5)

Ch1 Method : Confocal Fluorescence 405/488/561/640nm
Acquisition : BP447/50(Dual) Light Source : 405nm (30%)
Ch2 Method : Confocal Fluorescence 405/488/561/640nm
Acquisition : BP522/42(Dual) Light Source : 488nm (30%)
Ch3 Method: Confocal Fluorescence 405/488/561/640nm
Acquisition: BP600/37 Light Source: 561nm (30%)
Ch4 Method: Confocal Fluorescence 405/488/561/640nm
Acquisition: BP676/29 Light Source: 640nm (30%)

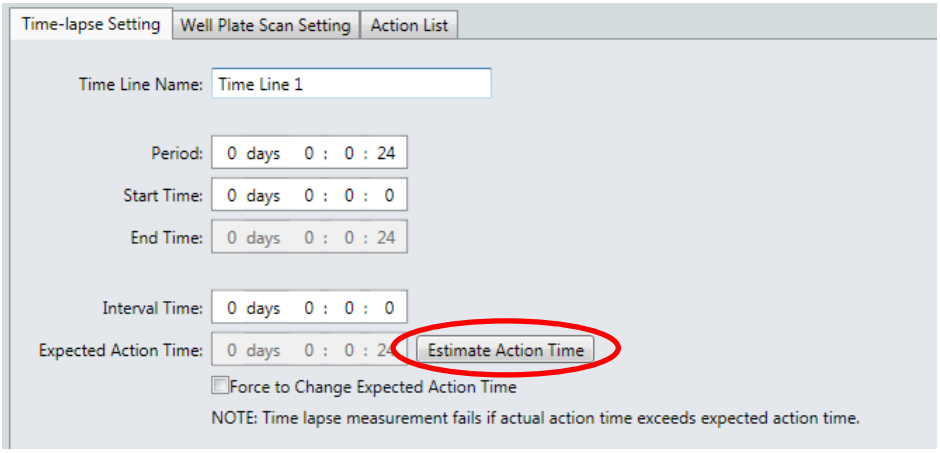


4) Set the items on the Action List tab. (Refer to 5.7)
Software focus is applied to Ch1. Images are captured for Ch1, Ch2, Ch3 and Ch4 on the software focus plane. To capture images on the software focus plane, set “0” under “Z Offset” for “Fluorescence Acquisition.”



5) Set the imaging wells and view field on the Well Plate Scan Setting tab. (Refer to 5.6)

6) Click “Estimate Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3)



7) Save the measurement setting file. (Refer to 5.11)

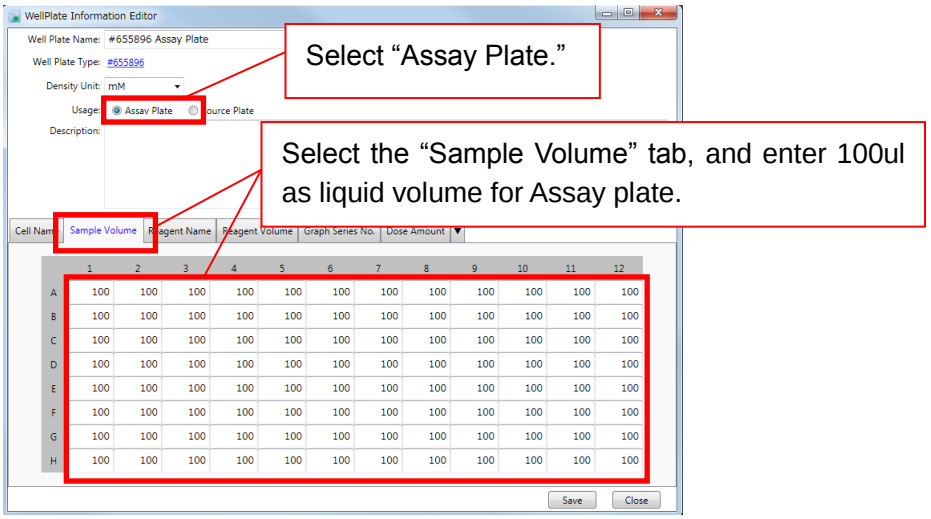
7.12. Example of Dispensing Setting

This section explains about commonly-used setting for well plate information files of Assay plate and Source plate (refer to 4.1), and dispensing setting file (refer to 5.12) to perform measurement using dispensing by 96-well tip rack.

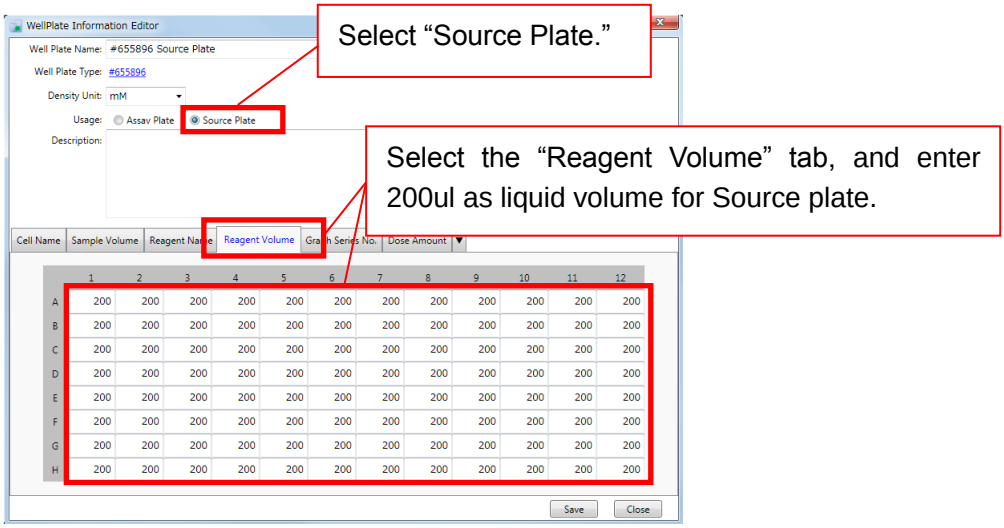
Setting #1 (To dispense below liquid surface of Assay plate)

Dispense 50ul to Assay plate from Source plate in which liquid amount of 200ul is added each well. Dispensing is performed from tip top at the Z position which moved to 1000um below from the liquid surface of Assay plate. This setting is the most common use.

1) Create each of wellplate information files for Assay plate and Source plate. (Refer to 4.1.)



Setting for Assay plate

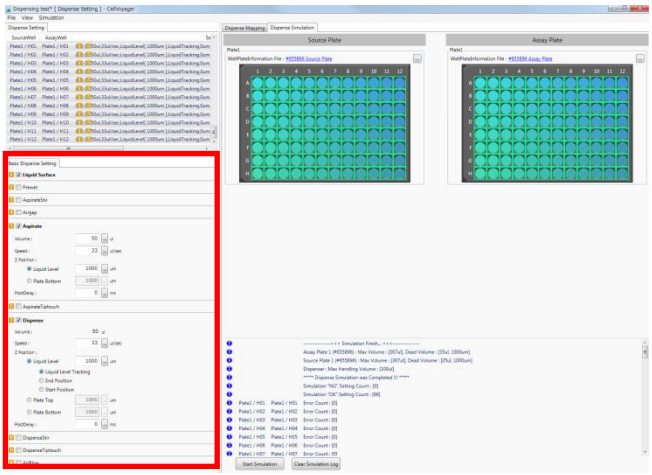


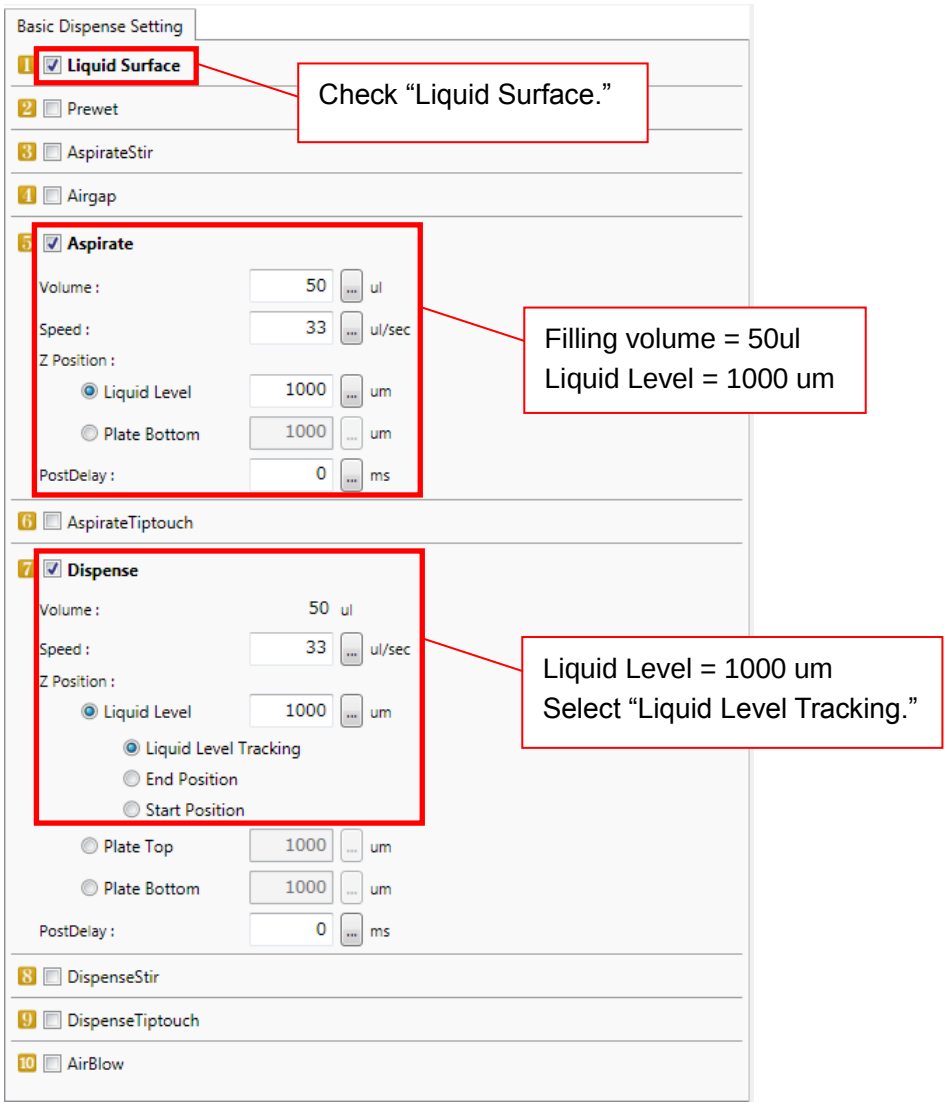
Setting for Source plate

MEMO

●Indication of reagent volume is 20% - 40% of well volume in Assay Plate and 50% - 80% of well volume in Source Plate.

2) Create a dispensing setting file. (Refer to 5.12.)

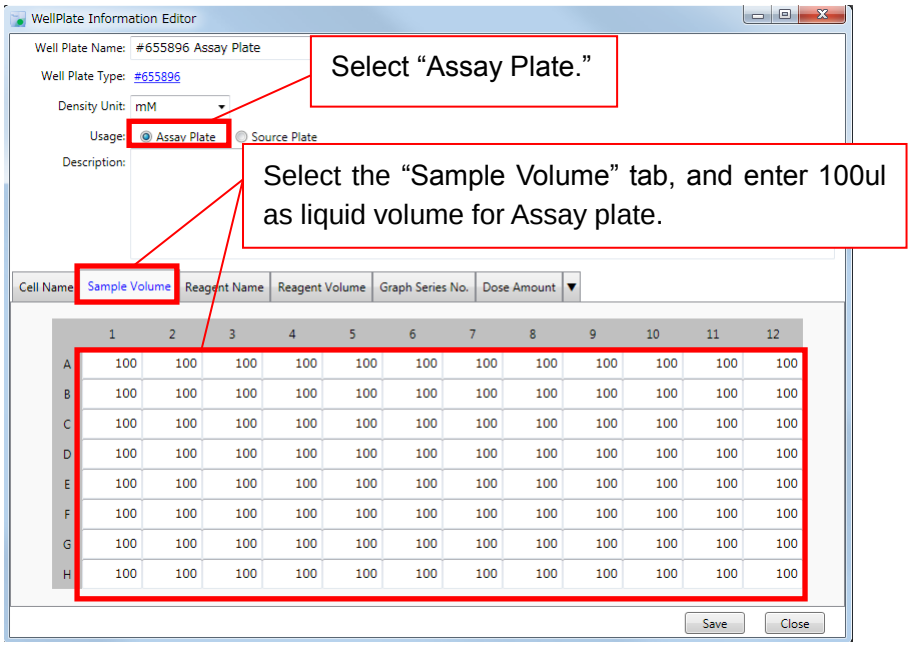




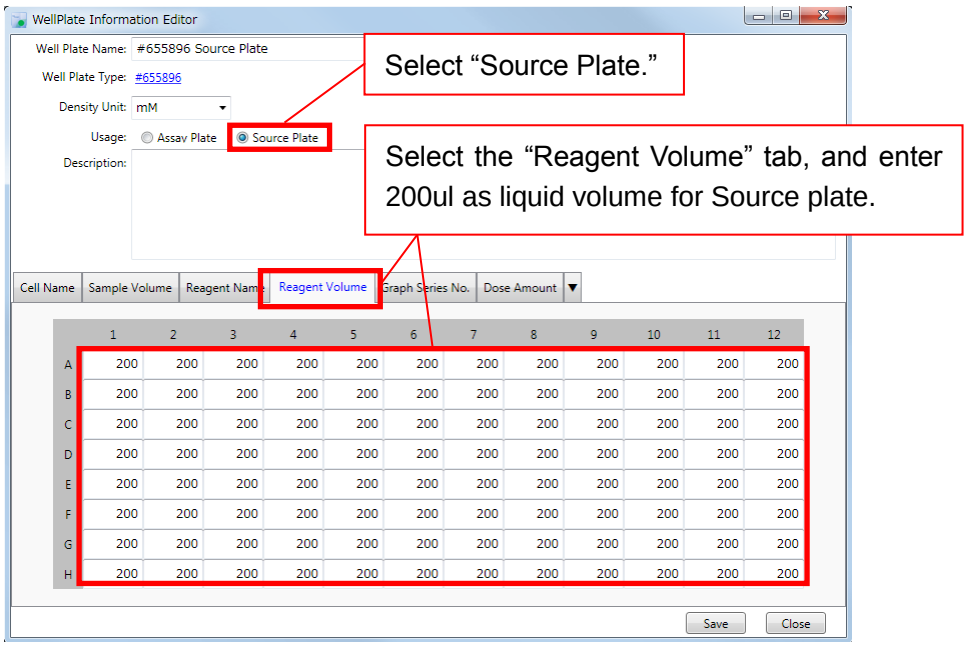
Setting #2 (To dispense above liquid surface of Assay plate)

Dispense 50ul to Assay plate from Source plate in which liquid amount of 200ul is added each well. Dispensing is performed from tip top at the Z position which moved to 2000um above from the liquid surface of Assay plate.

1) Create each of wellplate information files for Assay plate and Source plate. (Refer to 4.1.)

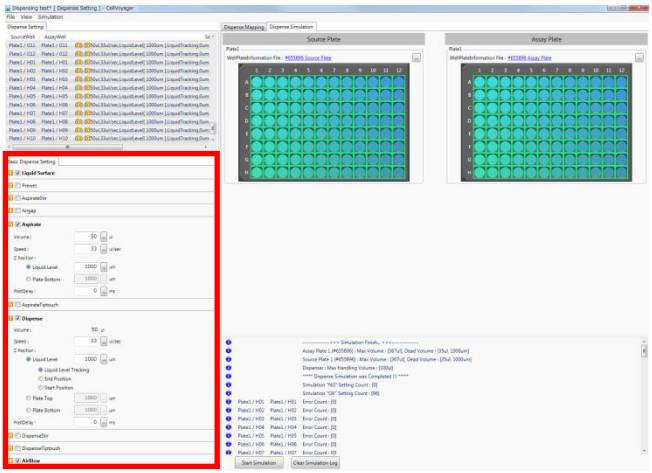


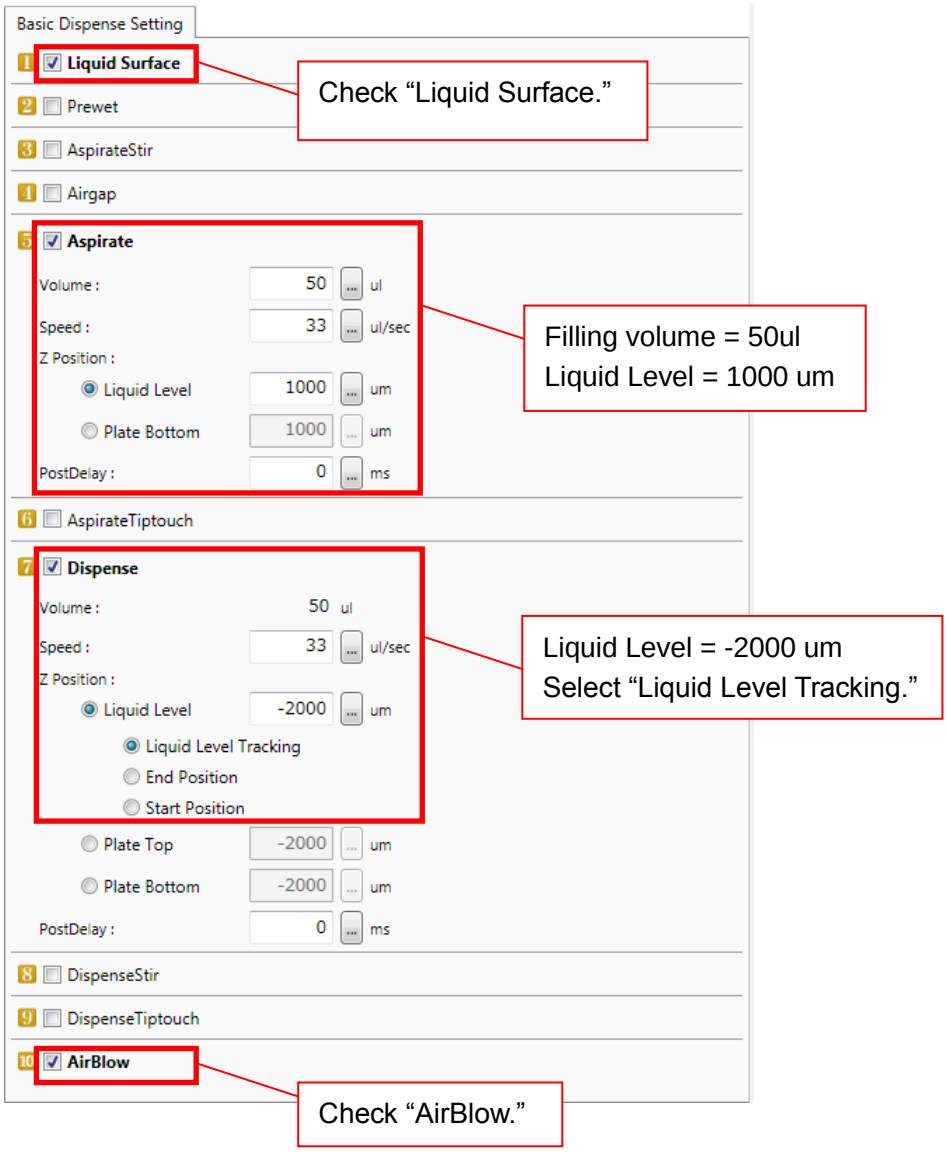
Setting for Assay plate



Setting for Source plate

2) Create a dispensing setting file. (Refer to 5.12.)

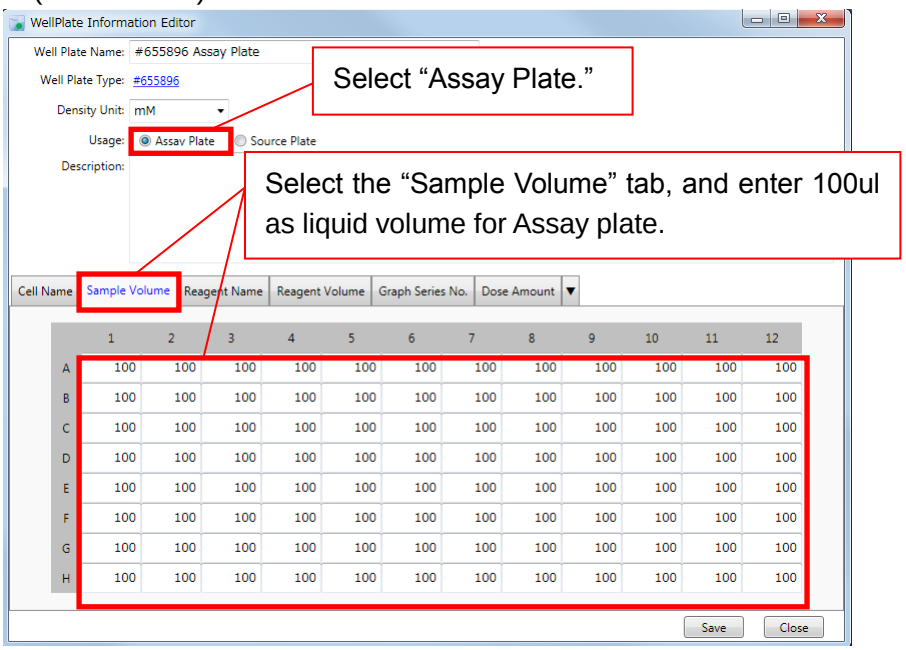




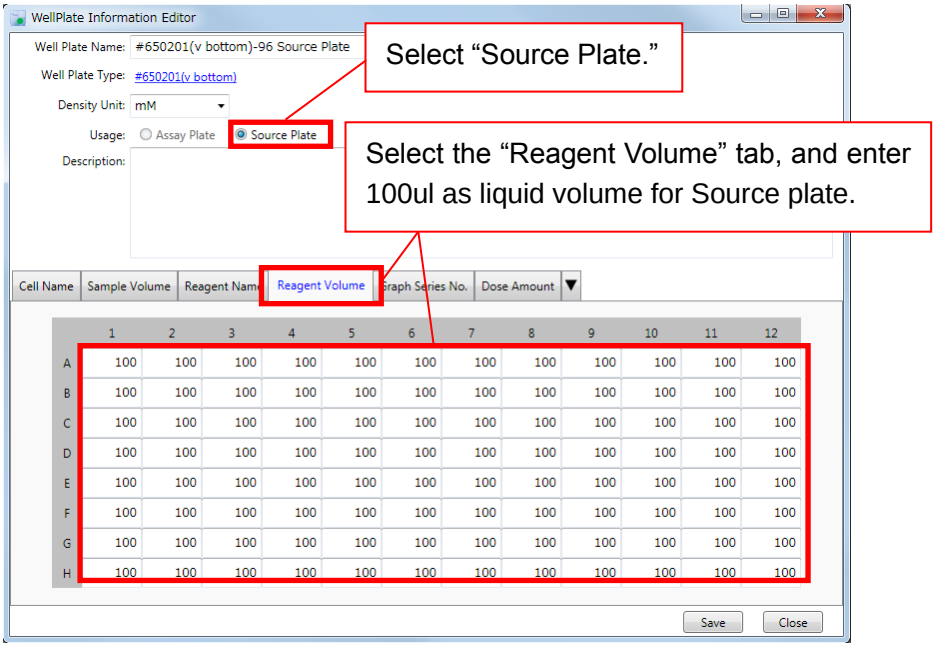
Setting #3 (To use liquid volume of Source plate until around Dead Volume)

Dispense 80ul to Assay plate from V-bottom Source plate (Dead Volume: 8ul) in which liquid amount of 100ul is added each well.
Dead Volume indicates the minimum volume so as not to make contacts between the tip top and the bottom surface of Source plate.

1) Create each of wellplate information files for Assay plate and Source plate. (Refer to 4.1.)

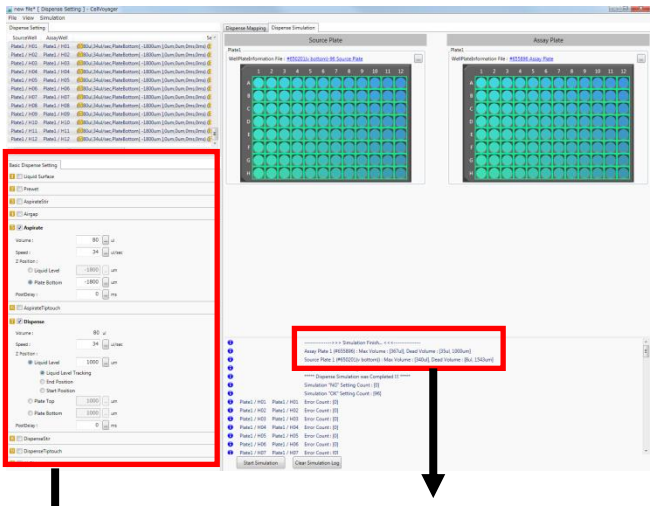


Setting for Assay plate



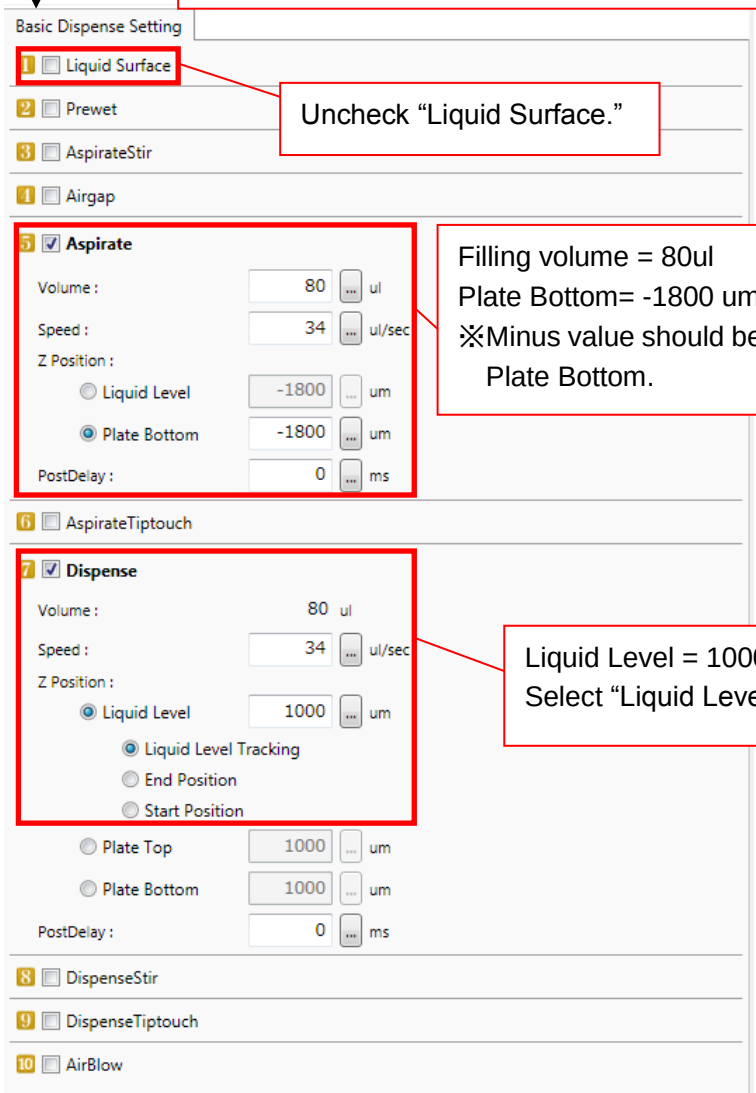
Setting for Source plate

2) Create a dispensing setting file. (Refer to 5.12.)



Assay Plate 1 (#655896) : Max Volume : [367ul], Dead Volume : [35ul, 1000um]
Source Plate 1 (#650201(v bottom)) : Max Volume : [340ul], Dead Volume : [8ul, 1543um]

Dead Volume of Source plate (Volume: 8ul, Height: 1543um)
※Dead Volume is different each of plate type. So, it can be confirmed by dispensing simulation



Uncheck “Liquid Surface.”

Filling volume = 80ul
Plate Bottom= -1800 um
※Minus value should be entered for Plate Bottom.

Liquid Level = 1000 um
Select “Liquid Level Tracking.”

MEMO

- To use solution until around Dead Volume, it is recommended to use the plates which has V-shaped bottom.
- Refer to the following table to confirm Dead Volume of each plate type.

Indication for Dead Volume of each plate type

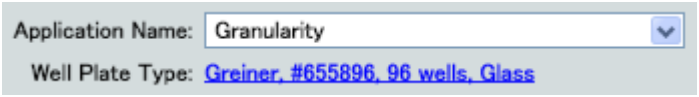
Well number/ Well shape	Well bottom shape	Source plate Dead Volume
96-well / Round-shaped	Flat	Almost 100ul
384-well / Square-shaped	Flat	Almost 25ul
96-well / Round-shaped	V-bottom / Round-shaped	Almost 10ul
384-well / Square-shaped	V-bottom / Round-shaped	Almost 10ul
384-well / Square-shaped	V-bottom / Square-shaped	Almost 10ul

7.13. High-Speed Time-Lapse Imaging with Multi Dispensing

Hitg-speed time-lapse imaging is performed with a laser beam of 488nm in wavelength at an imaging interval of 400ms and imaging time of 70s per well, with dispensing performed 5s after the start of imaging. Furthermore, another compound is dispensed 35s after the start of imaging in a same well.

1) Open the measurement setting file edit screen. (Refer to 5.2)

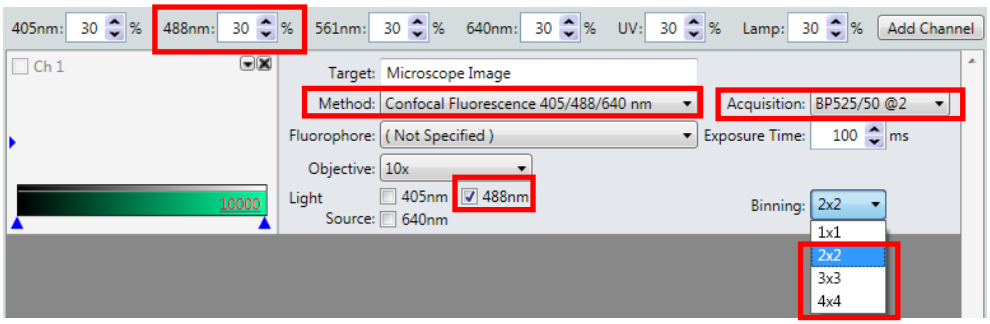
2) Enter the application name. (Refer to 5.4)



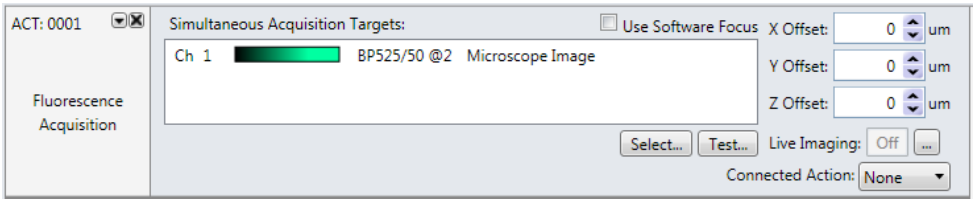
3) Set the imaging channels. (Refer to 5.5)
Ch1 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP525/50 Light Source : 488nm (30%) Binning: 2

MEMO

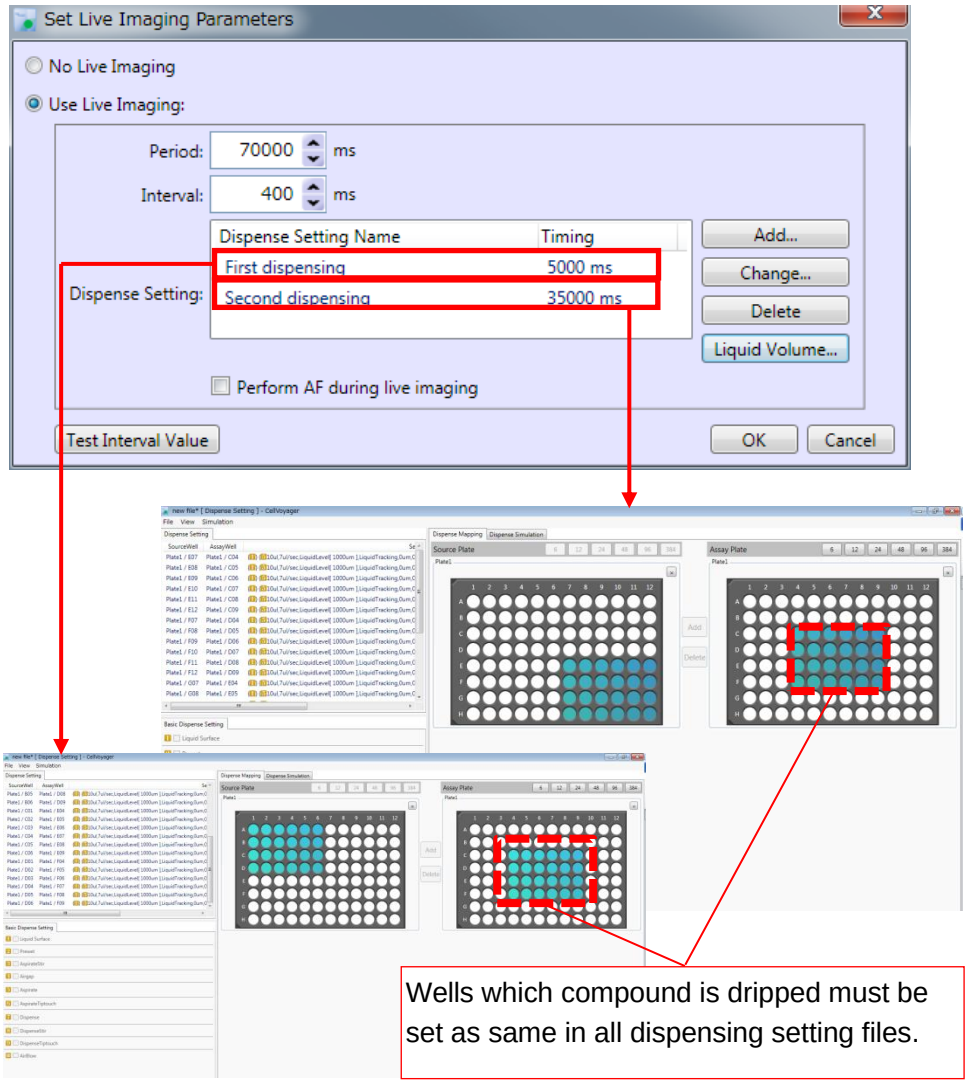
●Since a very large amount of image data will be acquired in high-speed time-lapse imaging, the binning setting more than “2x2” is recommended.



4) Set the items on the Action List tab. (Refer to 5.7)
To perform fluorescence imaging, set “Fluorescence Acquisition” task.

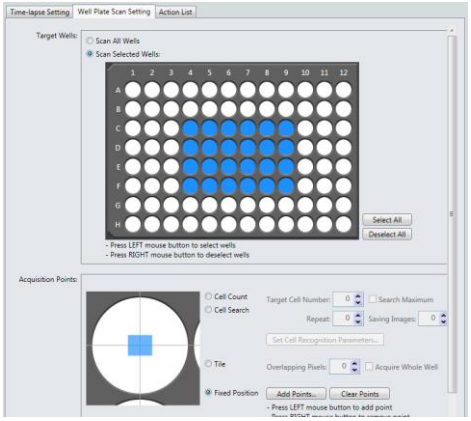


5) Set high-speed time-lapse imaging. (Refer to 5.8)
Up to 3 dispensing setting files (refer to 5.12, 7.12) can be assigned.



The first dispensing is performed from upper left 24wells of Source Plate to center 24wells of Assay Plate 5s after starting imaging.
The second dispensing is performed from lower right 24wells of Source Plate to center 24wells of Assay Plate 35s after starting imaging.
An imaging interval is 400ms. Time-lapse imaging finishes 70s after staring imaging and move to next well.

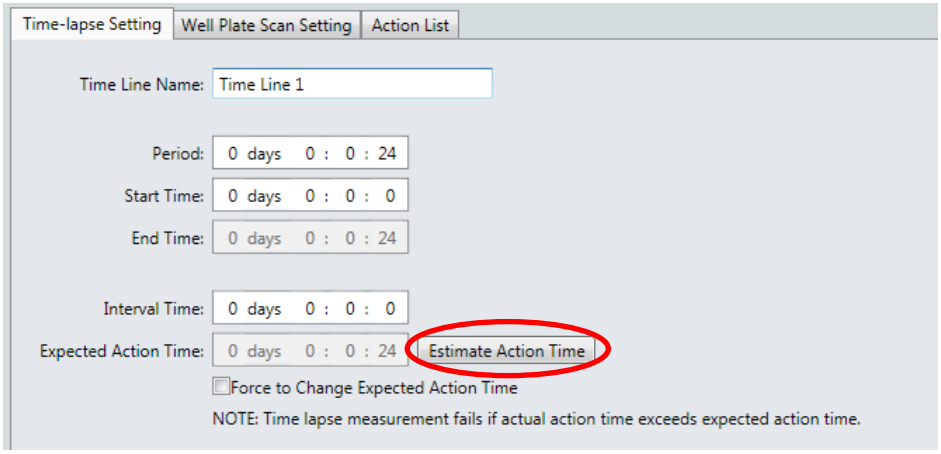
6) Set the imaging wells and view field on the Well Plate Scan Setting tab. (Refer to 5.6)



MEMO

- When images are captured by high-speed time-lapse imaging, the view field covers only one point at the center of the well.
- Select the same wells which is assigned in well-plate map of Assay Plate in dispensing setting file (refer to

7) Click “Estimate Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3)



8) Save the measurement setting file. (Refer to 5.11)

7.14. Tile Imaging

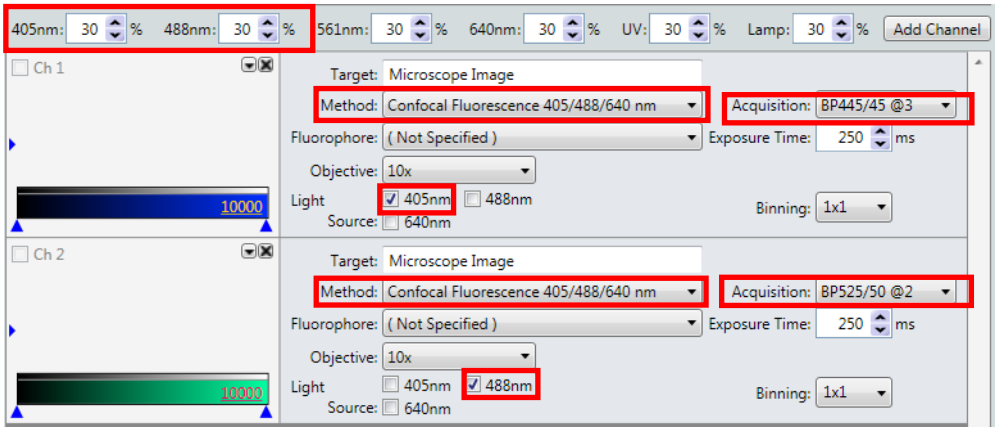
Image the whole/ partial region(s) of well as tile. Tiled image can be analyzed with displaying whole image. (Only Analysis Software supported model)

1) Open the measurement setting file edit screen. (Refer to 5.2)

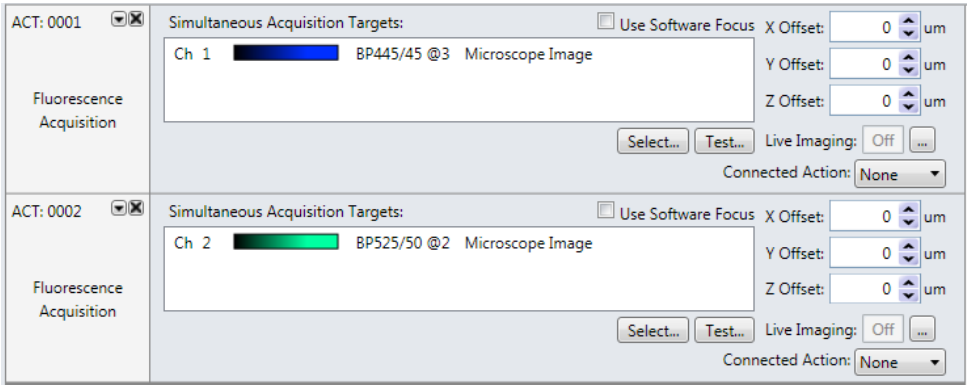
2) Enter the application name. (Refer to 5.4)

3) Set the imaging channels. (Refer to 5.5)

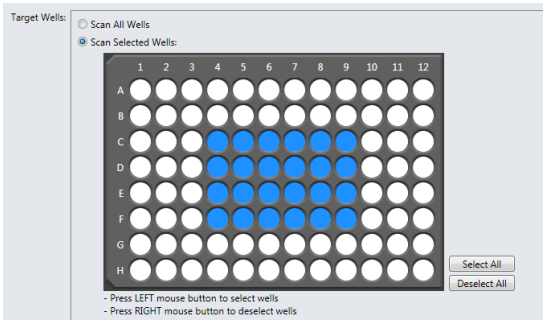
Ch1 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP445/45 Light Source : 405nm (30%)
Ch2 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP525/50 Light Source : 488nm (30%)



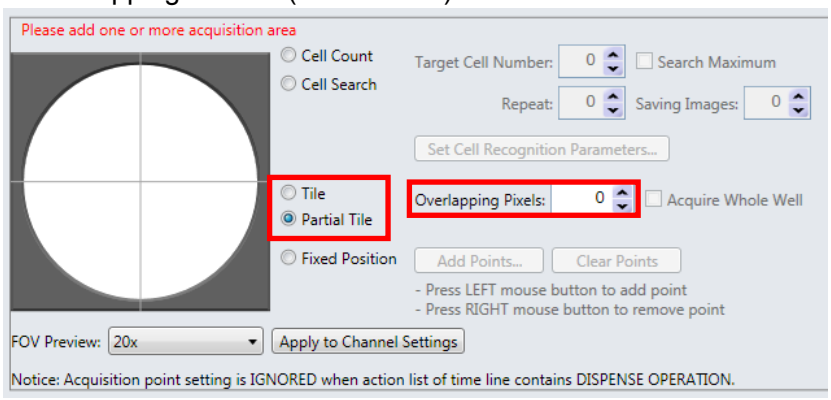
4) Set the items on the Action List tab. (Refer to 5.7)



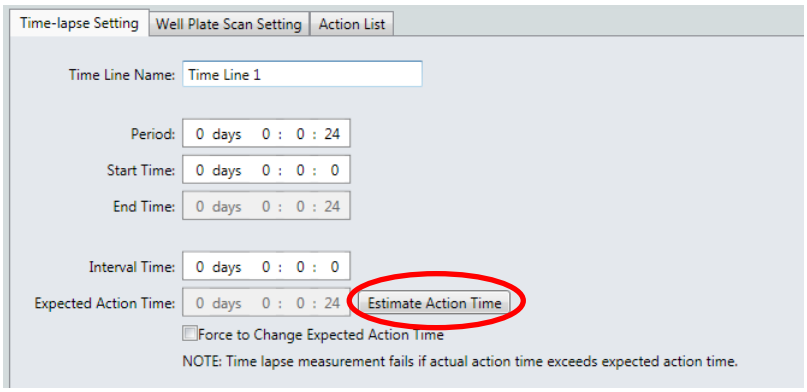
5) Set the imaging wells and view field on the Well Plate Scan Setting tab. (Refer to 5.6)



6) Select “Tile” or “Partial Tile” in Acquisition Points and assign the value of “Overlapping Pixels”. (Refer to 5.6)



7) Click “Estimete Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3)



8) Save the measurement setting file. (Refer to 5.11)

7.15. High-speed Time-lapse Imaging working with normal Imaging

After the one time imaging of DAPI stained nuclei by 405 nm laser, High-speed time-lapse imaging is performed by 488 nm laser at an imaging interval of 400 ms and imaging time of 30 seconds per well, with dispensing performed five seconds after the start of imaging.
High-speed Time-lapse Imaging only in the channel which is reactive to dispensing can reduce the amount of image data.

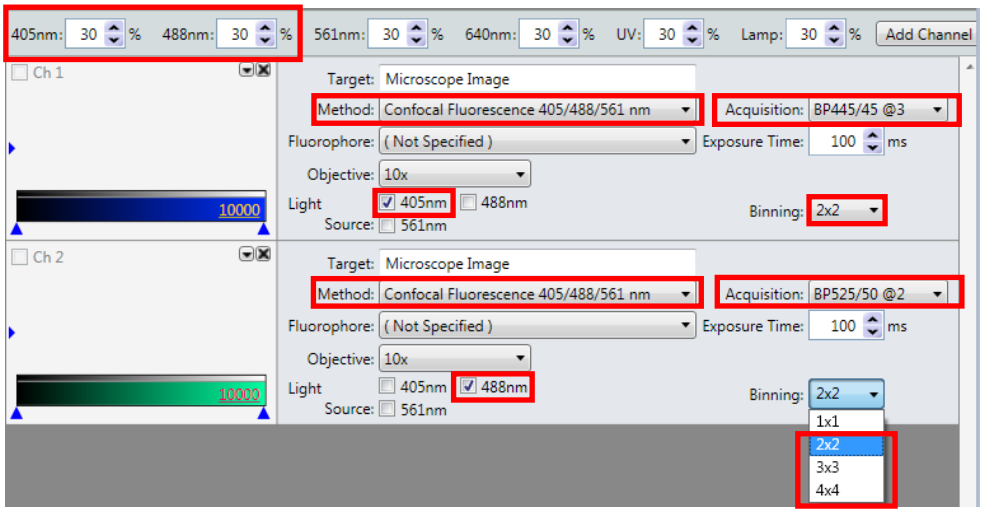
1) Open the measurement setting file edit screen. (Refer to 5.2)

2) Enter the application name. (Refer to 5.4)

3) Set the imaging channels. (Refer to 5.5)
Ch1 Method : Confocal Fluorescence 405/488/561nm
Acquisition : BP445/45 Light Source : 405nm (30%) Binning: 2
Ch2 Method : Confocal Fluorescence 405/488/561nm
Acquisition : BP525/50 Light Source : 488nm (30%) Binning: 2

MEMO

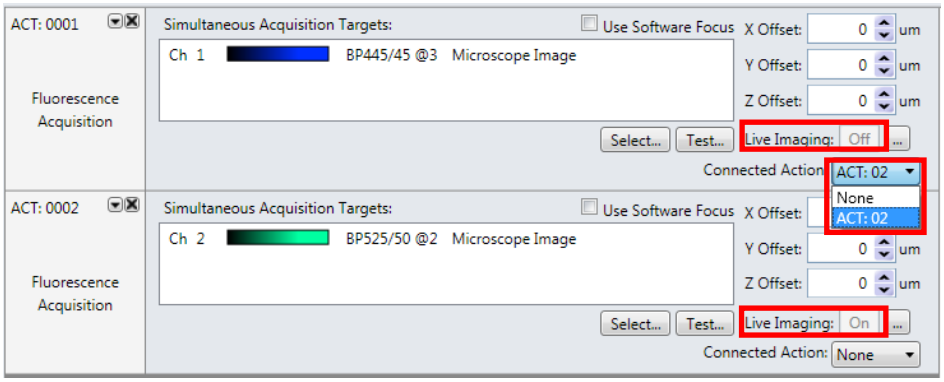
● Since a very large amount of image data will be acquired in high-speed time-lapse imaging, the binning setting more than “2x2” is recommended.



4) Set the items on the Action List tab. (Refer to 5.7)

To perform fluorescence imaging, set “Fluorescence Acquisition” task.

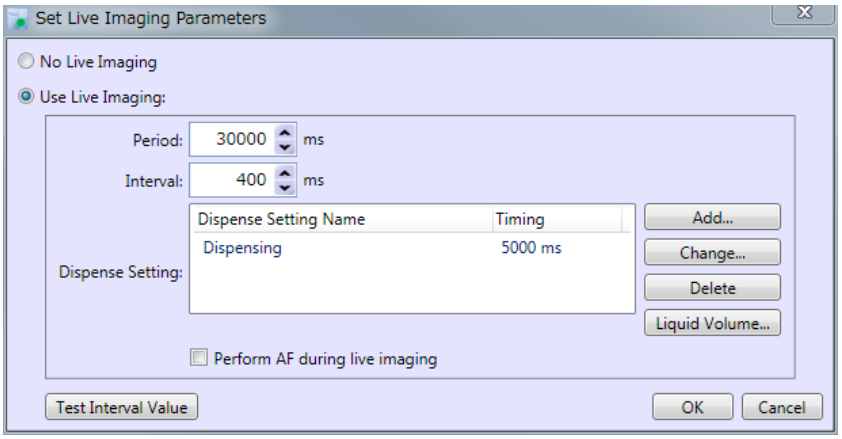
Set Live Imaging: ”On” only in ACT:0002, where Ch2 is set and set Connected Action:“ACT:02” in ACT:0001, where Ch1 is set. In this setting, imaging in Ch1 runs one time then High-speed time-lapse imaging in Ch2 starts consecutively. These images are linked as a set of data.



MEMO

- CV7000 Analysis Software can analyze these data.

5) Set high-speed time-lapse imaging. (Refer to 5.8)

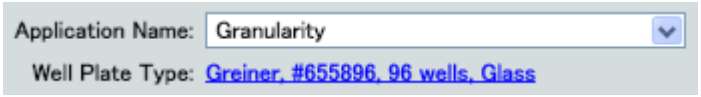


7.16. DPC Imaging

In Case of DPC Imaging Only

1) Open the measurement setting file edit screen. (Refer to 5.2)

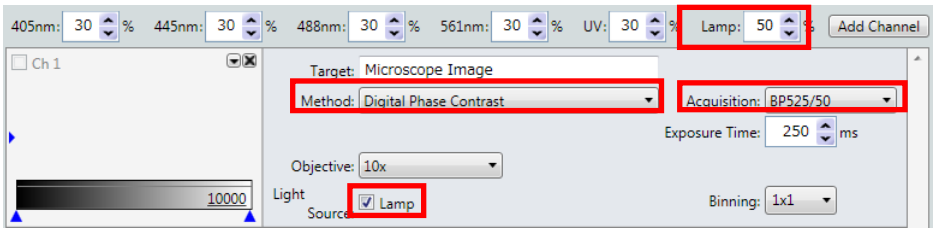
2) Enter the application name. (Refer to 5.4)



3) Set the imaging channels. (Refer to 5.5)

Ch1 Method : Digital Phase Contrast

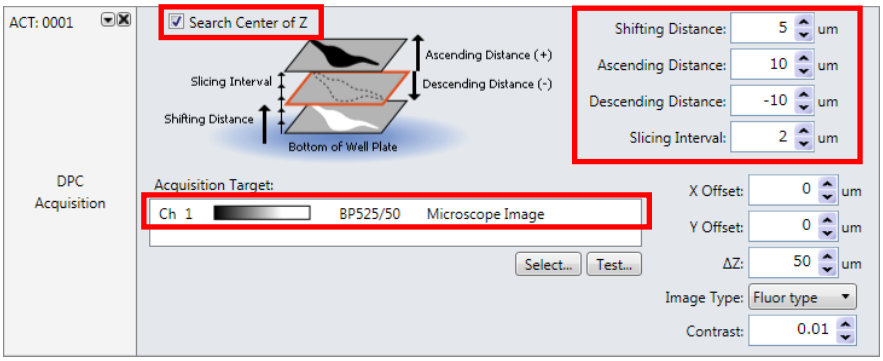
Acquisition : BP525/20 Light Source : Lamp (50%)



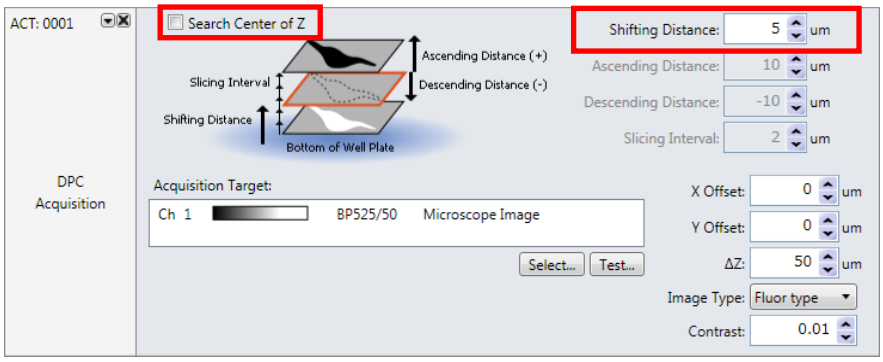
4) Set the item on the Action List tab. (Refer to 5.7)

DPC images are captured for Ch1.

In case of performing automatic DPC reference position search, check “Search Center of Z”.

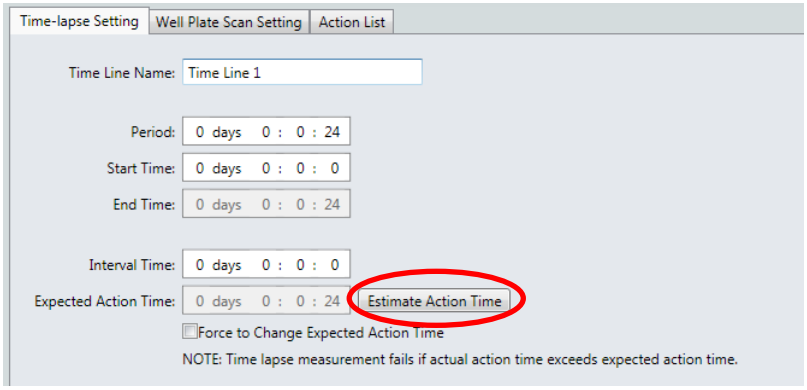


In case of without performing automatic DPC reference position search, uncheck “Search Center of Z”.



5) Set the imaging wells and view field on the Well Plate Scan Setting tab. (Refer to 5.6)

6) Click “Estimate Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3)



7) Save the measurement setting file. (Refer to 5.11)

Confocal Imaging and DPC Imaging of the Same View Field

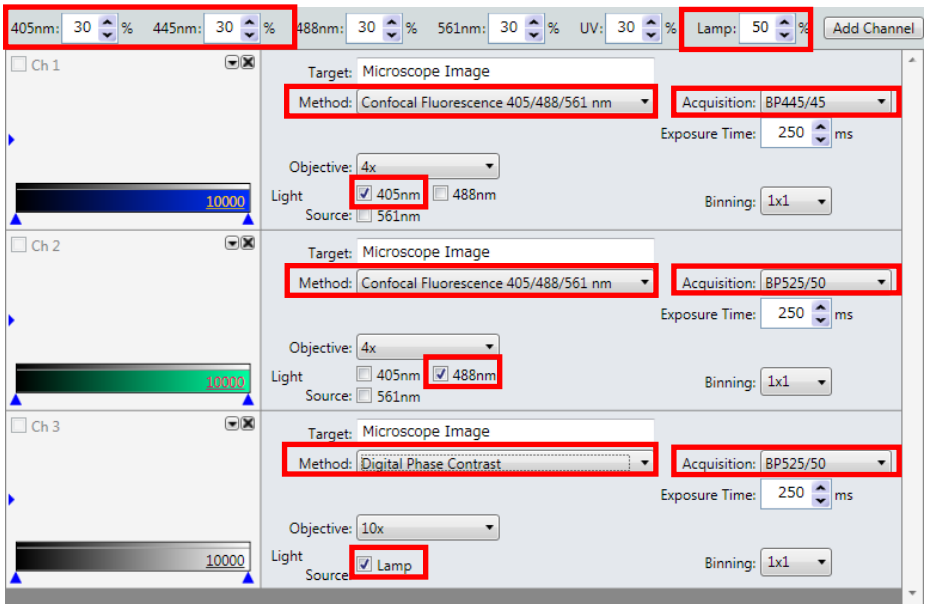
Confocal imaging is performed at two wavelengths (405 nm, 488 nm) and then imaging of the same view field is performed in the DPC mode.

1) Open the measurement setting file edit screen. (Refer to 5.2)

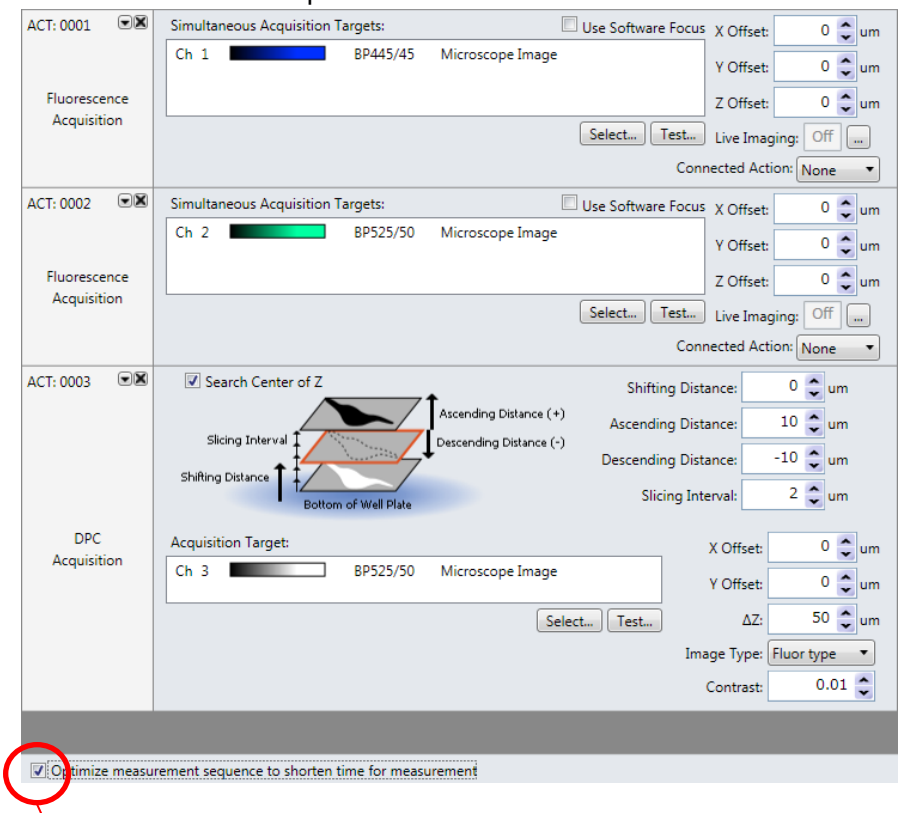
2) Enter the application name. (Refer to 5.4)

3) Set the imaging channels. (Refer to 5.5)

Ch1 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP445/45 Light Source : 405nm (30%)
Ch2 Method : Confocal Fluorescence 405/488/640nm
Acquisition : BP525/50 Light Source : 488nm (30%)
Ch3 Method : Digital Phase Contrast
Acquisition : BP525/20 Light Source : Lamp (50%)

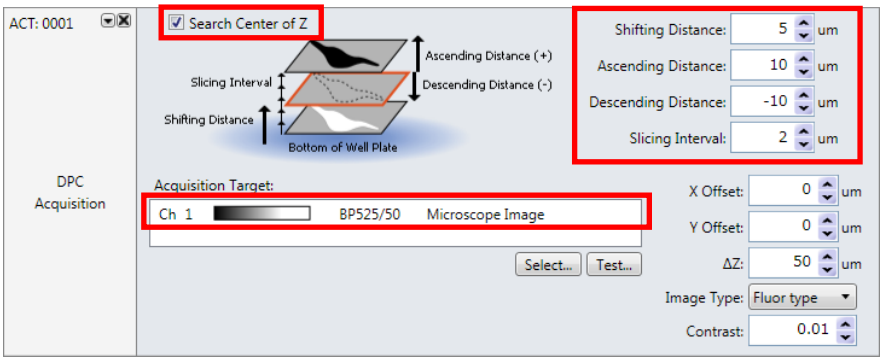


4) Set the items on the Action List tab. (Refer to 5.7)
Set Ch1 and Ch2 for the Fluorescence Acquisition tasks, respectively.
Set Ch3 for the DPC Acquisition task.

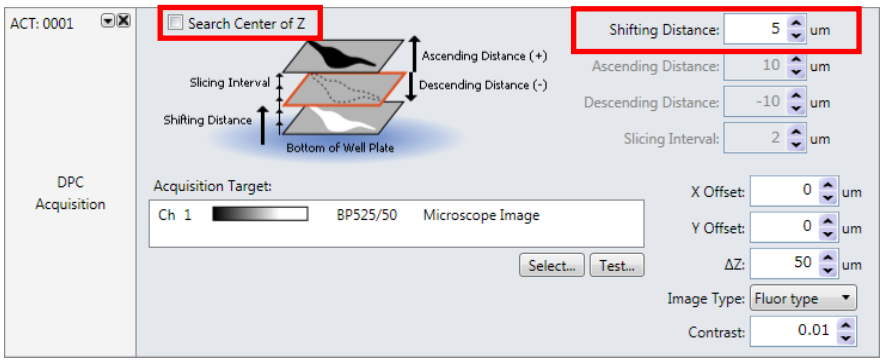


Check here to acquire hole well plate by the confocal fluorescence and then by bright field.
If the checkbox is unchecked, each image data is acquired at the same timing in the case that there are the differences of optical method and objective lens settings in the action list with switching optics..

In case of performing automatic DPC reference position search, check “Search Center of Z”.

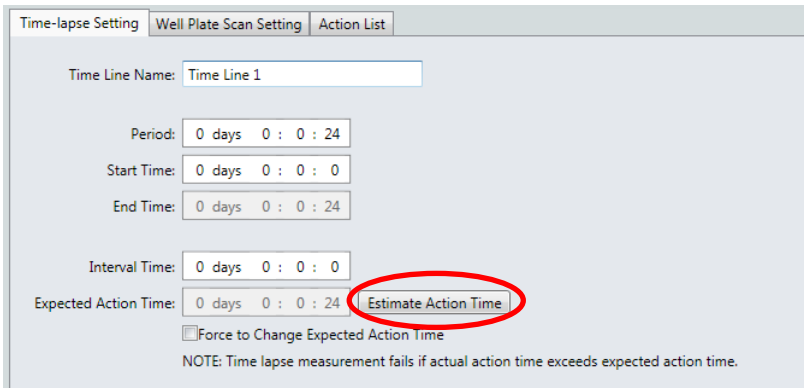


In case of without performing automatic DPC reference position search, uncheck “Search Center of Z”.



5) Set the imaging wells and view field on the Well Plate Scan Setting tab. (Refer to 5.6)

6) Click “Estimate Action Time” on the Time-lapse Setting tab to check the measurement time. (Refer to 5.3)



7) Save the measurement setting file. (Refer to 5.11)

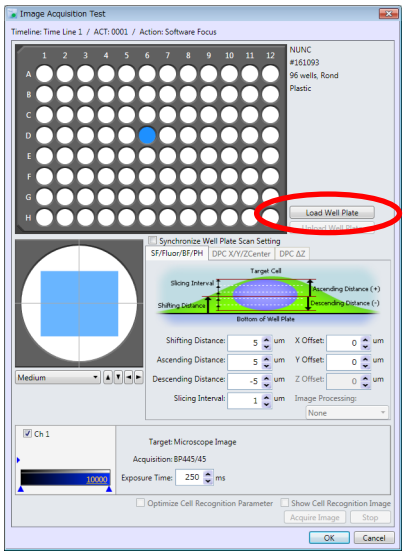
8. Measurement

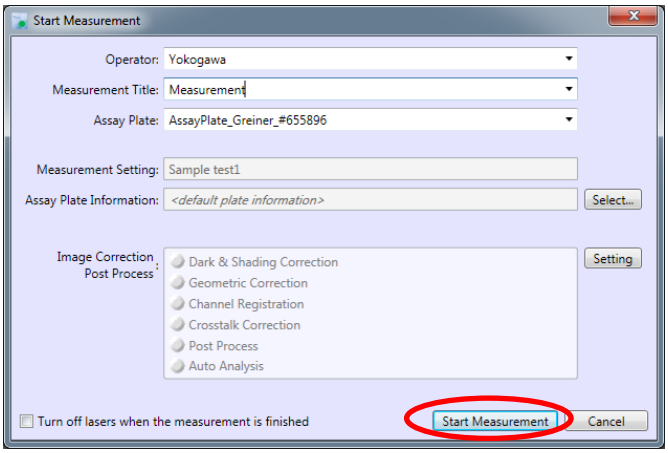
8.1. Setting Well Plates

⚠ WARNING

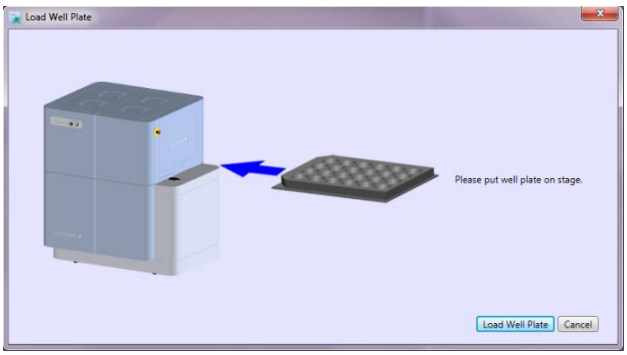
●DO NOT set well plate on lifter when stage is inside of CV7000. Well plate or stage can be injured.

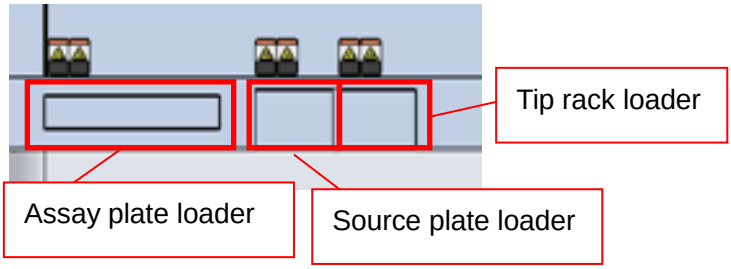
1) Set a well plate on the assay plate loader before displaying a measurement preview or starting measurement.
Click “Load Well Plate” on the Image Acquisition Test screen (refer to 5.9 and 6.2), or click “Start Measurement” on the Start Measurement screen (refer to 8.3 and 8.4).





2) The stage moves to the loader exit. When the stage has fully moved to the specified position, the Load Well Plate screen opens. (Do not click “Load Well Plate.”)

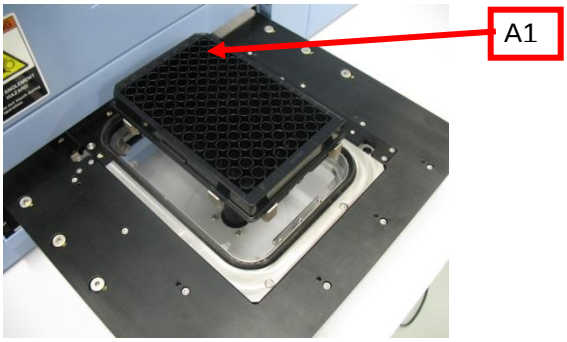




⚠ WARNING

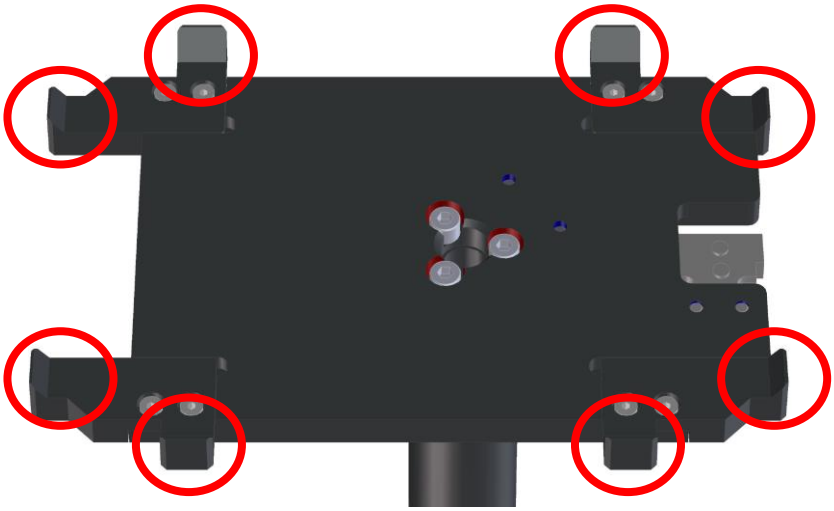
●DO NOT touch loader exit area while moving the loaders.

3) Set the assay plate on the stage. Set the plate so that the well “A1” on the well plate comes to the top left-hand corner of the stage.
In case of observation by slide glass, please refer to 8.7
(Only slide glass supported model)

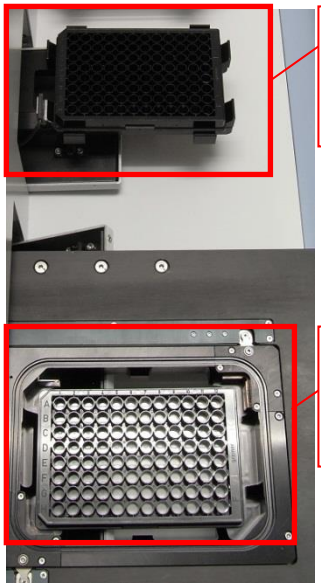


⚠ WARNING

●Set the assay plate so that its eight positions align with the eight positions of the stage. Failure to do so may cause measurement unit damage.



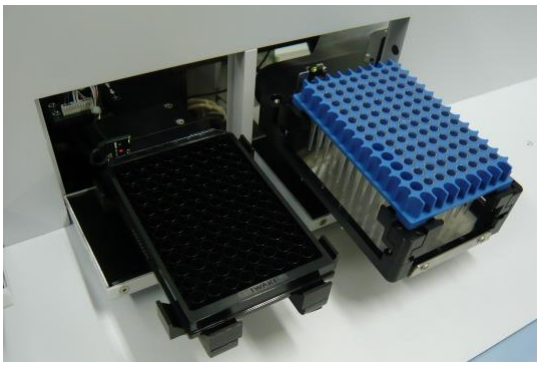
In the case of dispensing measurement, set the source plate on the shuttle at the back of the stage. Set the plate so that the well “A1” on the well plate comes to the top left-hand corner of the stage. In the case of dispensing measurement, be sure to remove the covers of the assay plate and source plate before setting the plates.

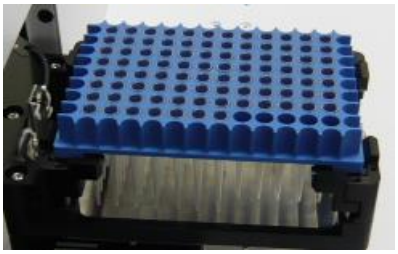


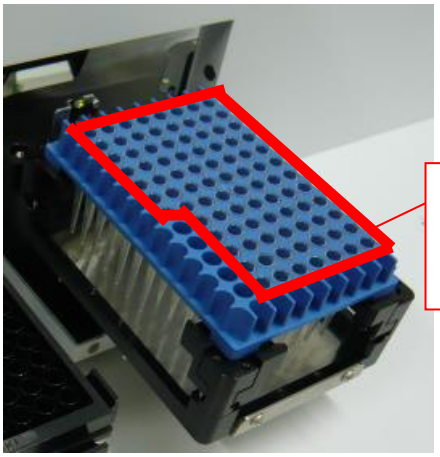
⚠ WARNING

- In the case of dispensing measurement, be sure to remove the covers of the assay plate and source plate before setting the plates. Failure to do so may cause dispenser damage.
- The Assay plate and Source plate simulated in the dispensing setting file (refer to 5.12) must be used in dispensing measurement. Failure to do so may cause dispenser damage.

4) In dispensing measurement, set a tip rack on the tip rack loader.
Dedicated tip rack for CV7000 must be used.
(200µl Nested Pipette Tips 105649 from Caliper Life Sciences is used for 96-tip rack model only.)
(LT-384-R from Corning is used for 384-tip rack model only.)





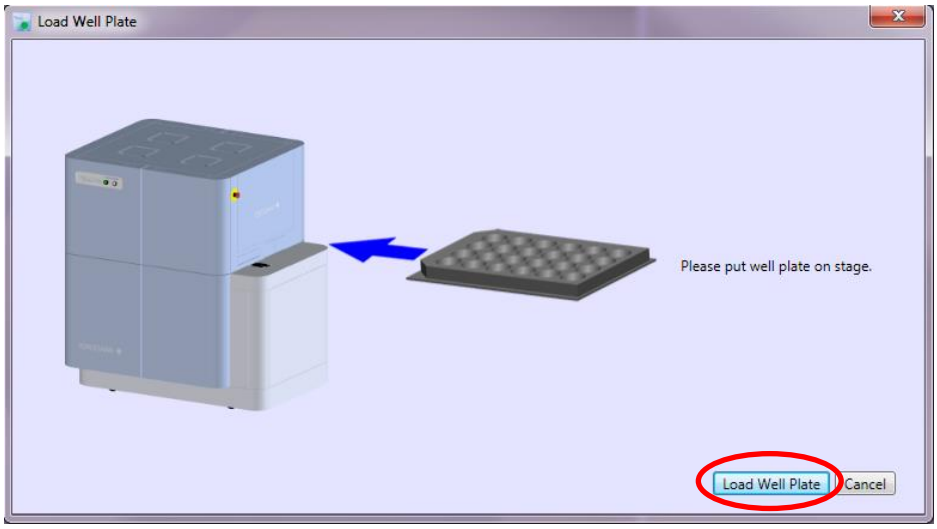


⚠ WARNING

- Be sure not to use 384-tip rack for 96-tip rack model. Failure to do so may cause dispenser damage.
- Be sure not to use 96-tip rack for 384-tip rack model. Failure to do so may cause dispenser damage.
- Make sure to use dedicated tip rack for each of 96-tip rack model and 384-tip rack model. Failure to do so may cause dispenser damage.

Put the tip disposal box beside CV7000.

5) After setting the plate, click “Load Well Plate” in the “Load Well Plate” screen to move the stage to the reader section.

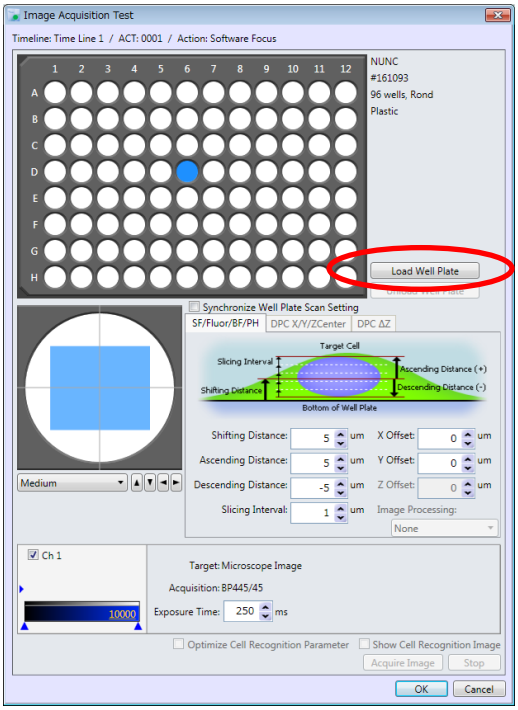


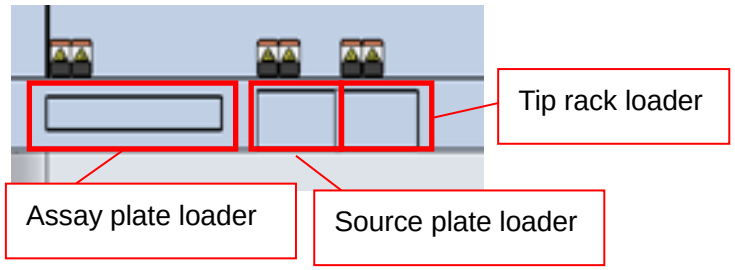
⚠ WARNING

●DO NOT touch loader exit area while moving the loaders.

8.2. Removing the Well Plates

1) When measurement of the assay plate is complete or “Unload Well Plate” on the Image Acquisition Test screen (refer to 5.9 and 6.2) is clicked, the plate is unloaded.

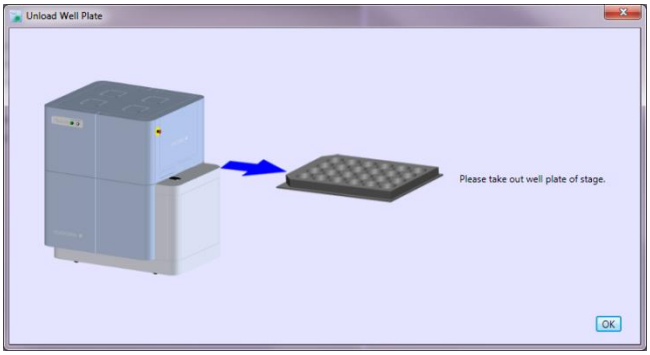




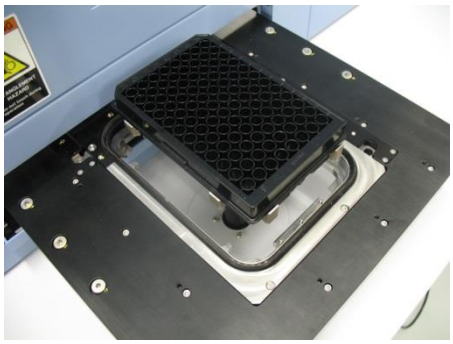
⚠ WARNING

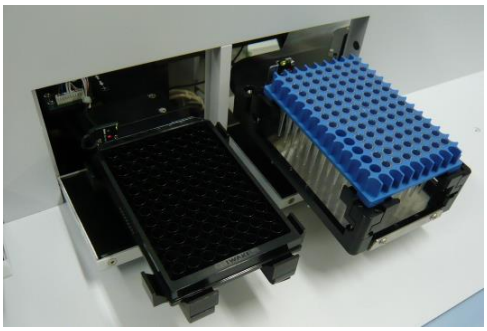
●DO NOT touch loader exit area while moving the loaders.

The Unload Well Plate screen opens. Do not click “OK.”

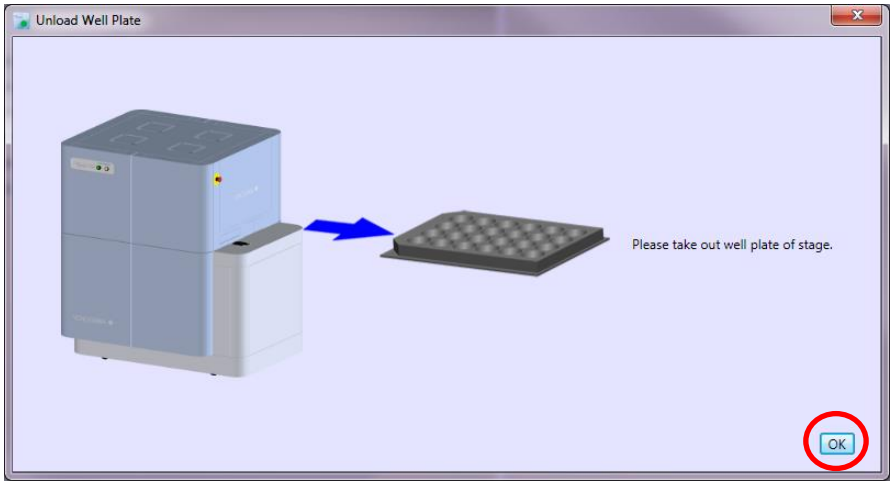


2) Remove the assay plate.
After dispensing measurement, remove also the source plate and tip rack.





3) After removing the plate, click “OK” on the Unload Well Plate screen.



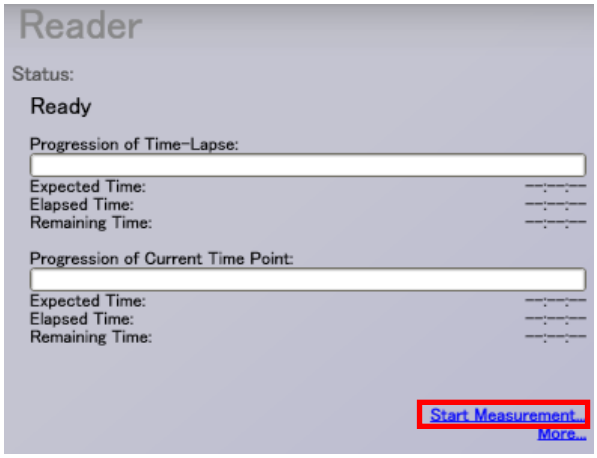
⚠ WARNING

●DO NOT touch loader exit area while moving the loaders.

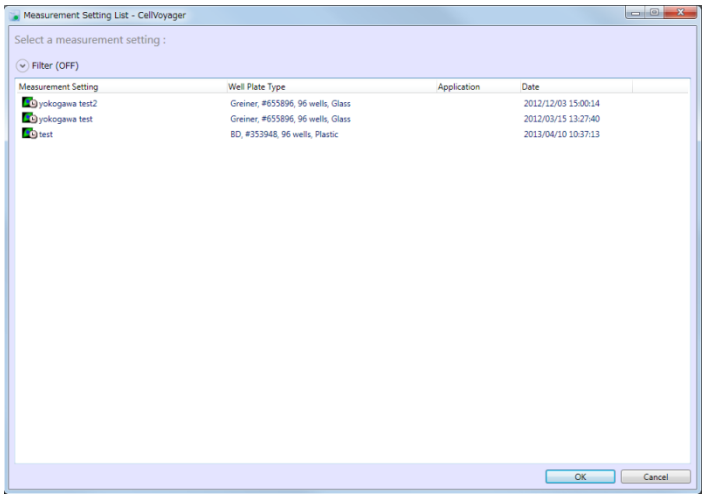
8.3. Measuring the Assay Plate without Dispensing

1) Create a measurement setting file and save the file. (Refer to Chapter 7)

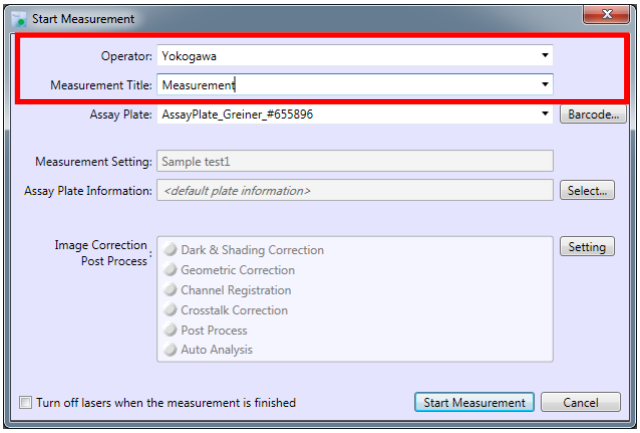
2) Click “Start Measurement” at the bottom of the Status section in the Reader area.



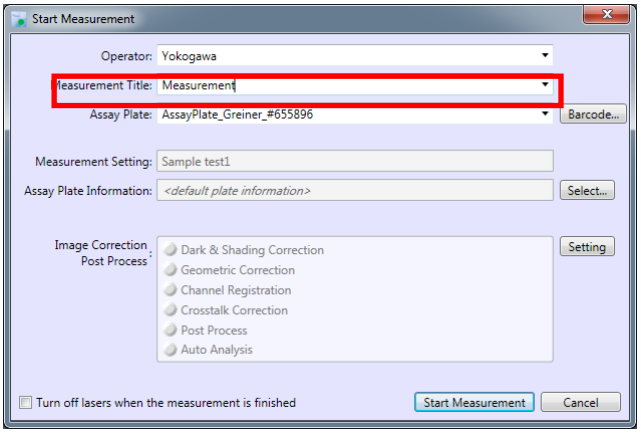
3) Select the measurement setting file. (Refer to 8.5.)



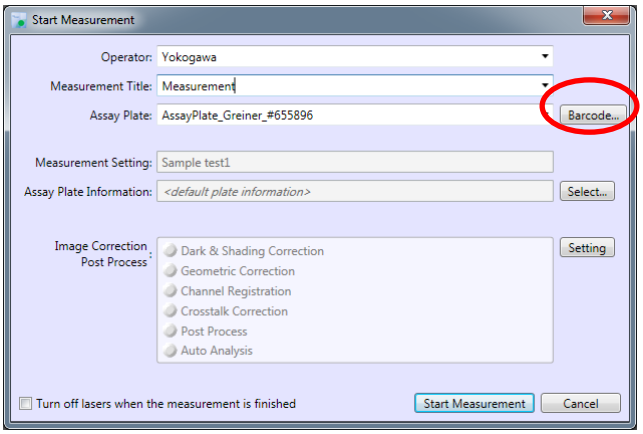
4) The Start Measurement screen opens. Enter appropriate text in the “Operator” and “Measurement Title” fields. The entered names will be used to generate the folder name for measured images. (Refer to 10.1)



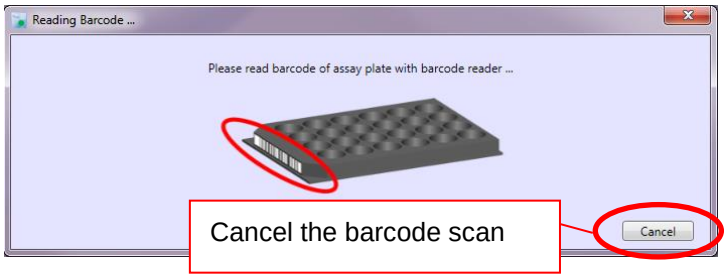
5) Enter appropriate text in the “Assay Plate” field for assay plate name. The entered names will be used to generate the folder names for measured images. (Refer to 10.1)



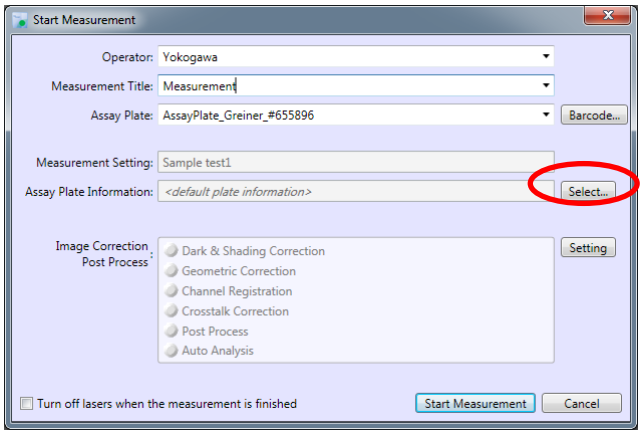
Click “Barcode” If there is a barcode on the assay plate.
(Barcode reader model only)



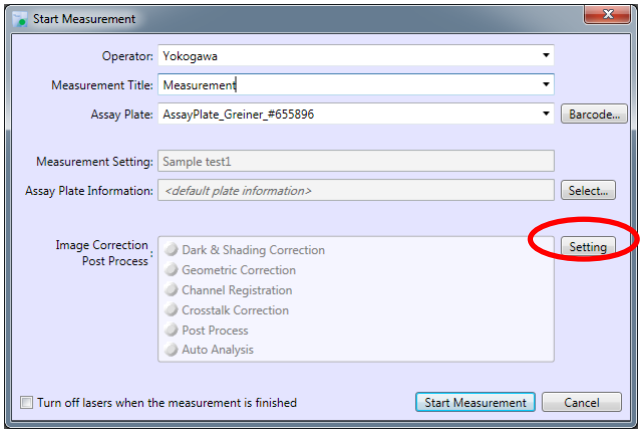
Barcode reader starts to scan. The scanned barcode data is entered to the “Assay Plate” field.



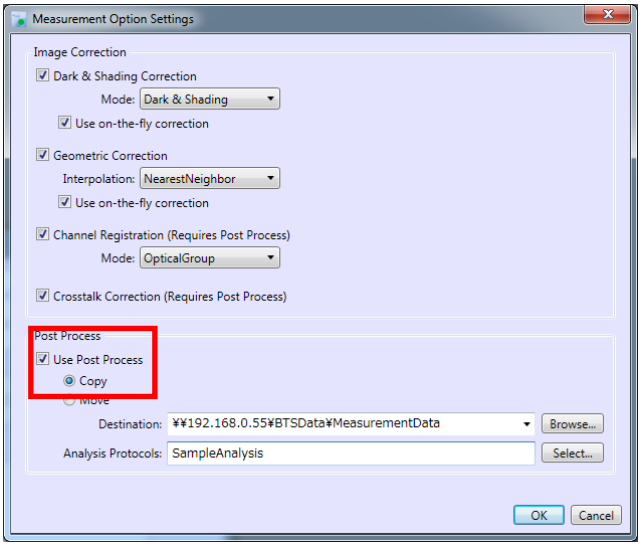
6) To specify a well plate information file for Assay plate, click “Select” and specify a desired file.
(Measurement can be performed with the default settings.)



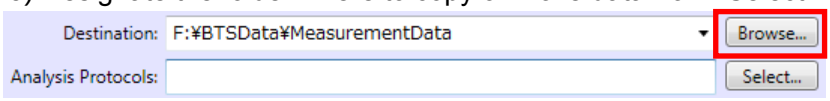
7) Click “Setting” if using post-processing function.



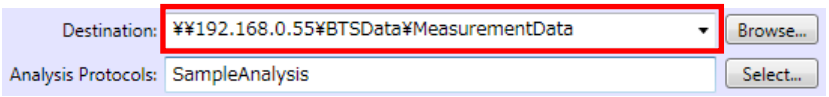
8) “Measurement Option Settings” window opens.
In case of using post-processing function, check “Use Post Process”.
Select “Copy” to automatically copy the measurement data to the designated folder. Select “Move” to automatically move the measurement data to the designated folder.



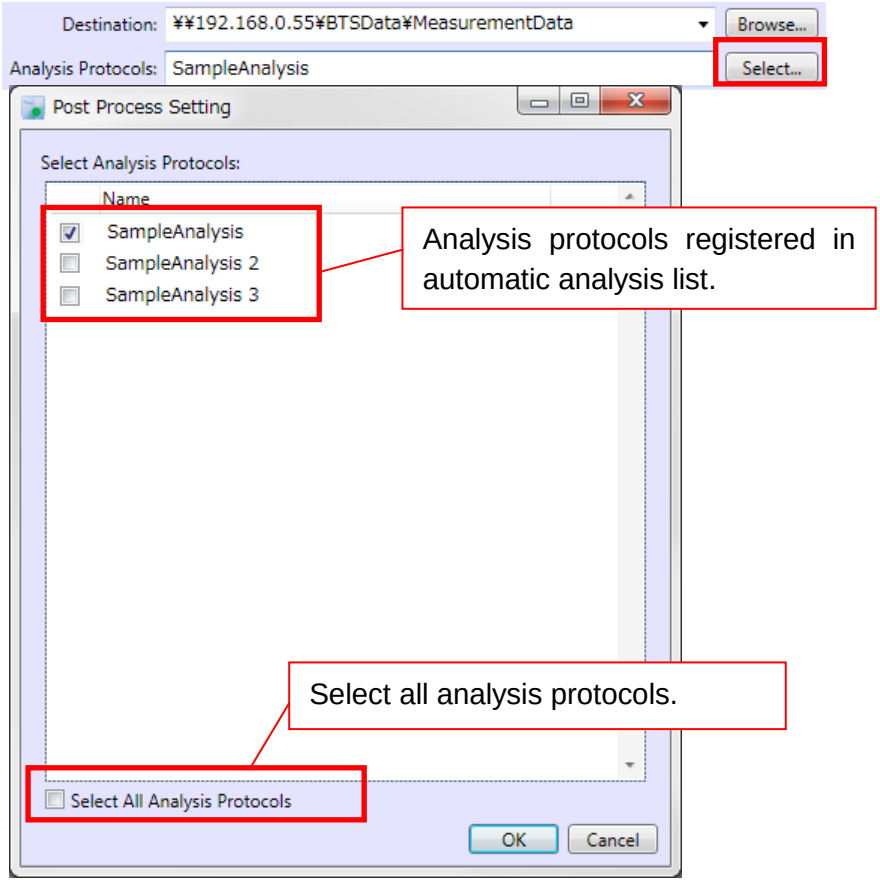
9) Designate the folder where to copy or move data from “Select”.



To perform automatic analysis, specify a folder selectable from CV7000 Analysis Software.
(Example: Specify “D:¥BTSData¥MeasurementData” in the WS for analysis as “Destination.”)



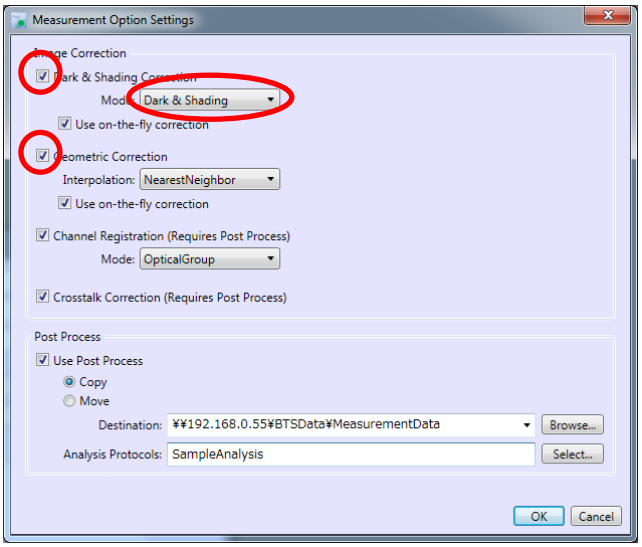
To perform automatic analysis, click “Select” of “Analysis Protocol” and then, select analysis protocols to be analyzed.
(About analysis protocol, refer to section 12.6 of IM 80H01C03-01E CV7000 Analysis Software User’s Manual)
Click “OK” after selecting.



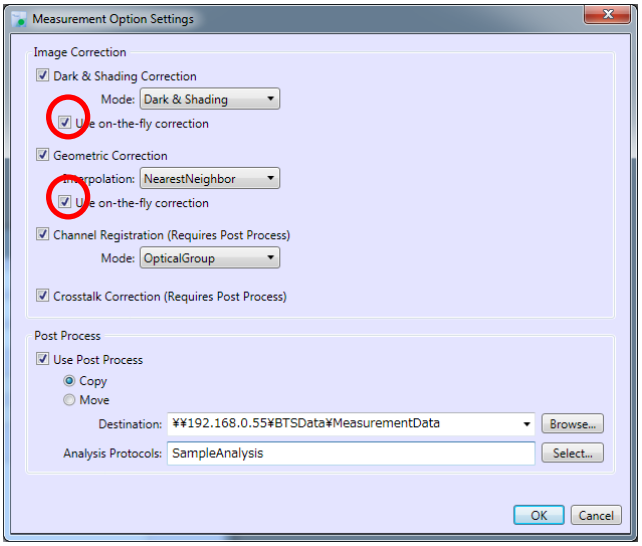
MEMO

●To display the AnalysisProtocol setting in the Start Measurement screen, launch the CV7000 Analysis Software and set to communicate the network previously.

10) Set image correction setting post imaging (for detail of image correction, please refer to IM 80H01A16-01E Image Correction Software User's Manual). Set "Dark & Shading Correction" and "Geometric Correction". "Dark & Shading" is recommended for "Dark & Shading Correction".

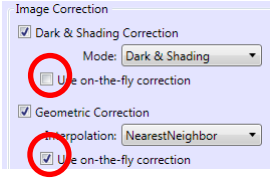


11) Check if performing on-the-fly correction.

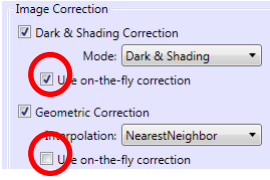


MEMO

- If "Use on-the-fly correction" is checked, image correction is performed before saving image to hard disk.
  Processing speed becomes faster than using Post Processor, but raw image data is not saved.
- It cannot to perform "Dark & Shading Correction" without using on-the-fly correction and perform "Geometric Correction" with using on-the-fly correction. (Opposite setting is available.)

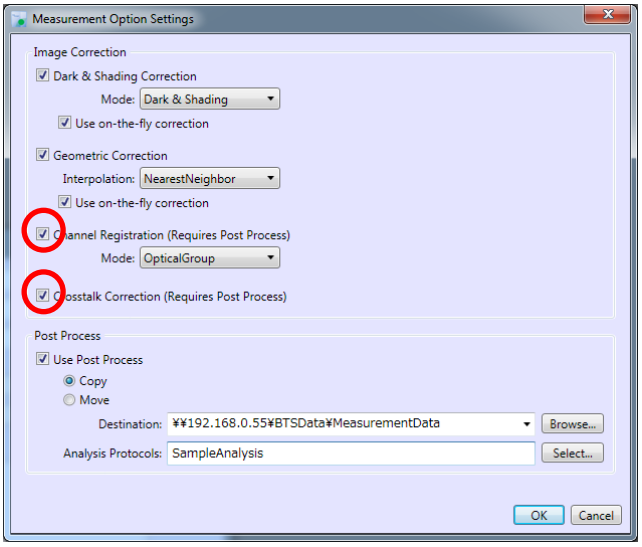


← NG



← OK

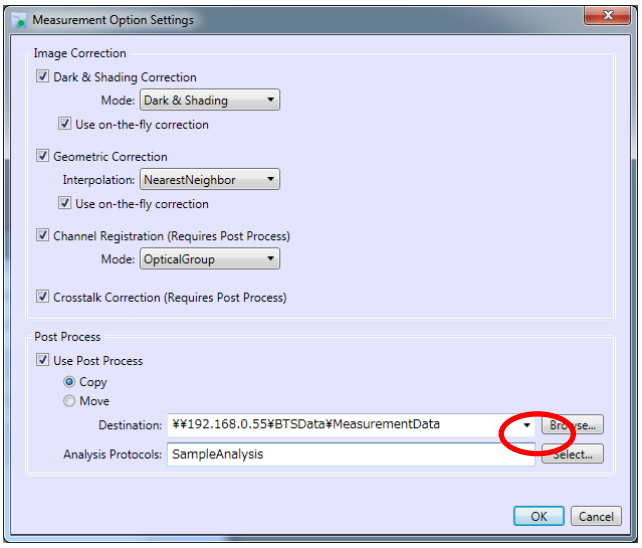
12) Set “Channel Registration” and “Crosstalk Correction”. These 2 items are selectable if “Use Post Process” is checked. Also, they are not correspond to on-the-fly correction.



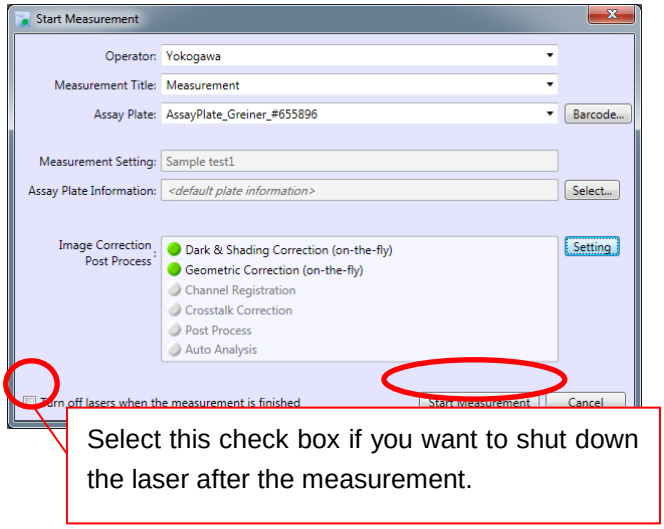
MEMO

- ●Don‘t select “Channel Registration” if the images are acquired by high-speed time-lapse mode.
- ●To correct fluorescent crosstalk, “Dark & Shading” must be selected in “Dark & Shading Correction”, and “Geometric Correction” or “Channel Registration” must be selected.
  It is recommended to select both “Geometric Correction” and “Channel Registration”.
- ●”Crosstalk Correction” cannot be performed if “Fluorophore” is not designated in Channel Setting (refer to 5.5)
- ●Emission spectrum of fluorophore can be deformed by surrounding environment (pH, temperature, saline concentration). In this case, crosstalk correction cannot be performed normally.
  Confirm image after crosstalk correction by Image Correction Software and if the images have abnormality, uncheck “Crosstalk Correction”.

13) Click “OK”.



14) Click “Start Measurement”.



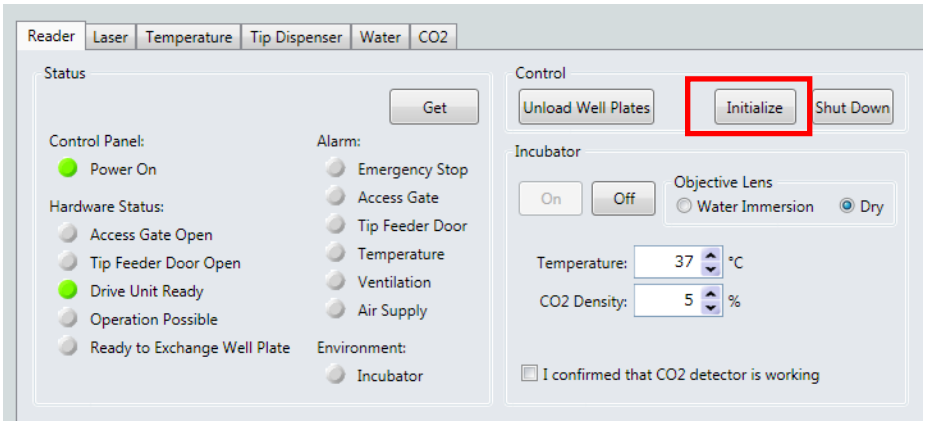
15) Set well plates on the sample loader. (Refer to 8.1)
Measurement will start once the plates are set.

16) When the measurement is complete, remove the well plates. (Refer to 8.2)

17) Measured results are displayed. (Refer to Chapter 9)

MEMO

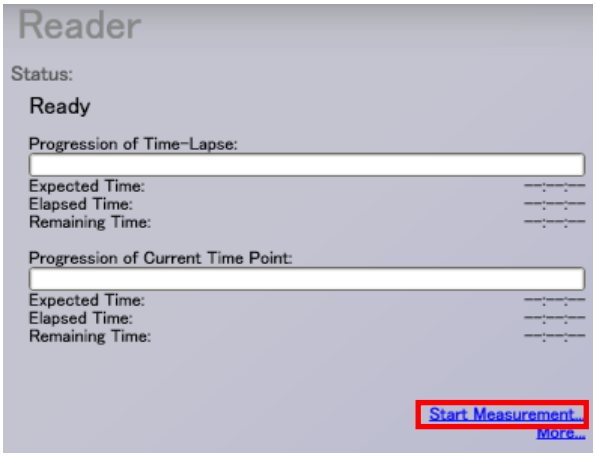
●To start the laser again, click “Initialize” on the Reader Status tab of the Reader Control screen.



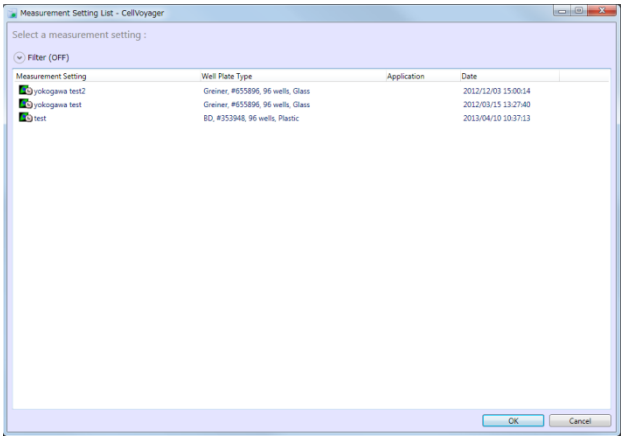
8.4. Measuring the Assay Plate with Dispensing

1) Create a measurement setting file and save the file. (Refer to Chapter 7)

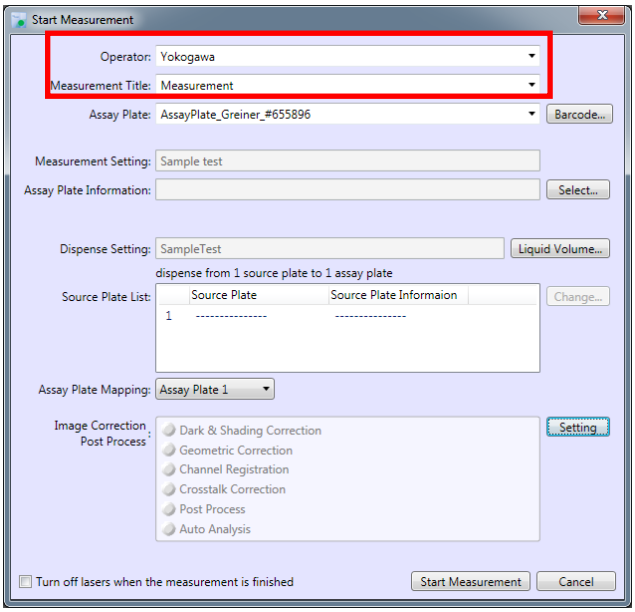
2) Click “Start Measurement” at the bottom of the Status section in the Reader area.



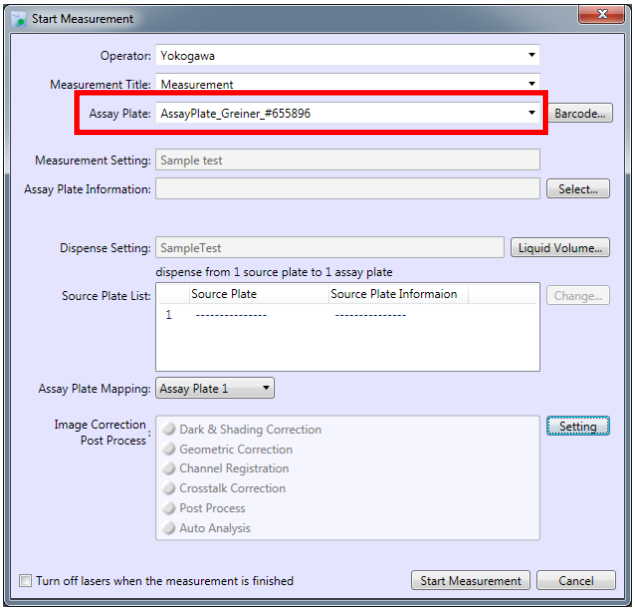
3) Select the measurement setting file. (Refer to 8.5.)



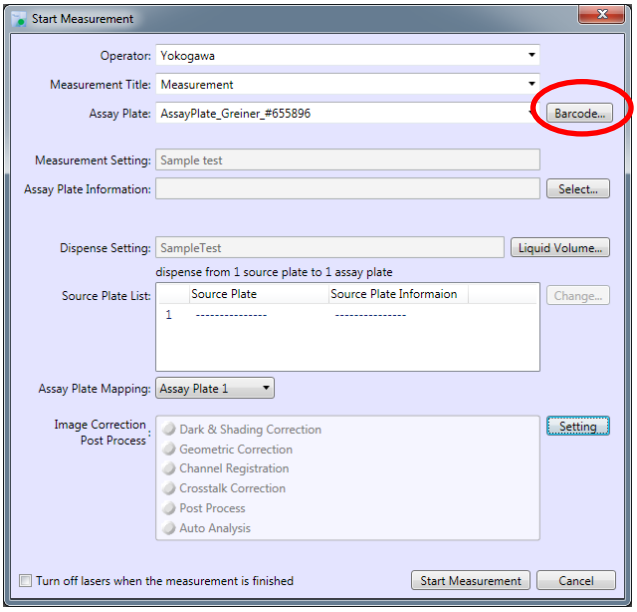
4) The Start Measurement screen opens. Enter appropriate text in the “Operator” and “Measurement Title” fields. The entered names will be used to generate the folder name for measured images. (Refer to 10.1)



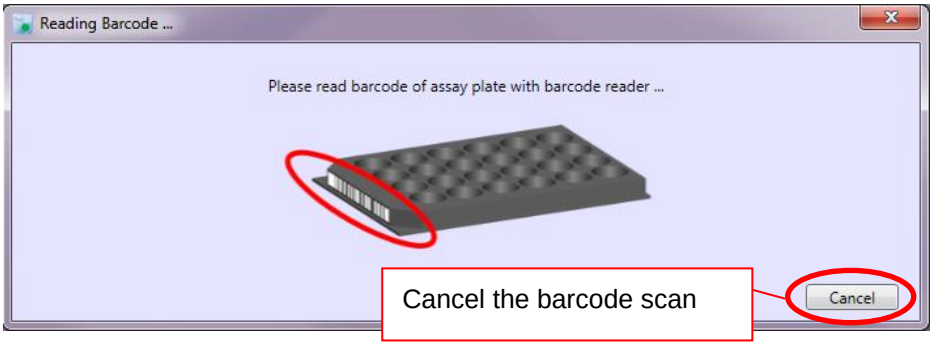
5) Enter appropriate text in the “Assay Plate” field for assay plate name. The entered names will be used to generate the folder name for measured images. (Refer to 10.1)



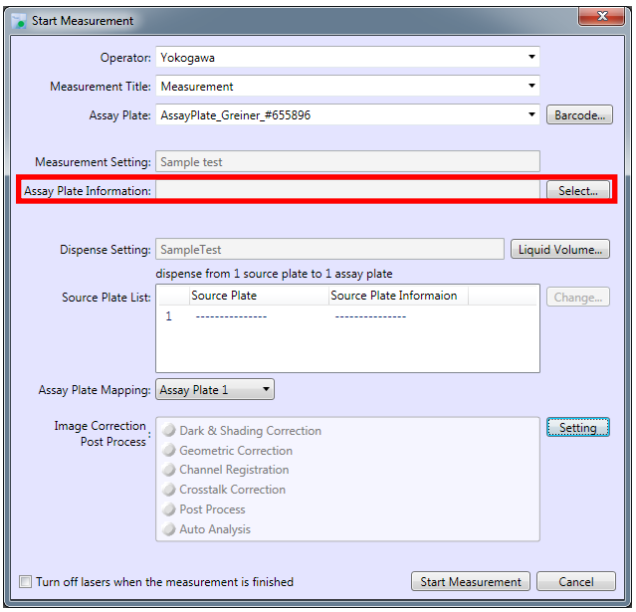
Click “Barcode” If there is a barcode on the assay plate.
(Barcode reader model only)



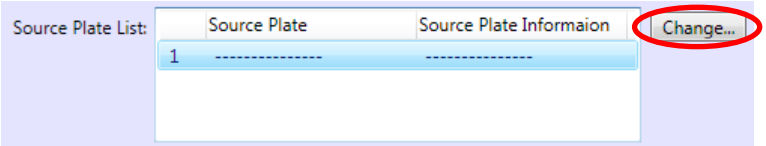
Barcode reader starts to scan. The scanned barcode data is entered to the “Assay Plate” field.



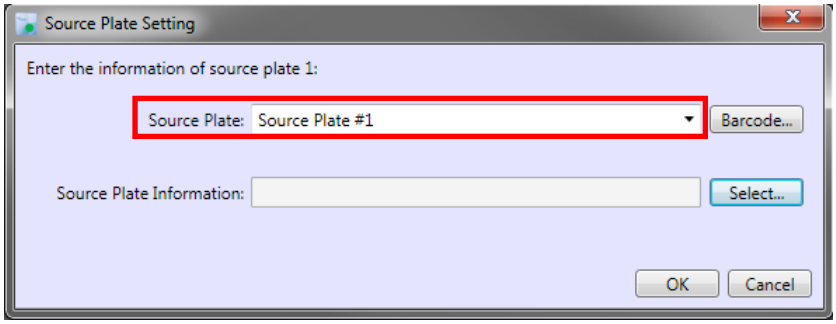
6) Click “Select” and select a desired well plate information file for the assay plate. (Refer to 4.1)



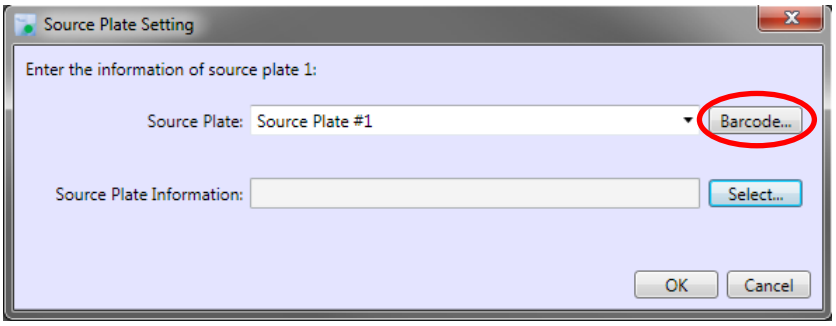
7) Click “Change” and select a well plate information file for the source plate. (Refer to 4.1)



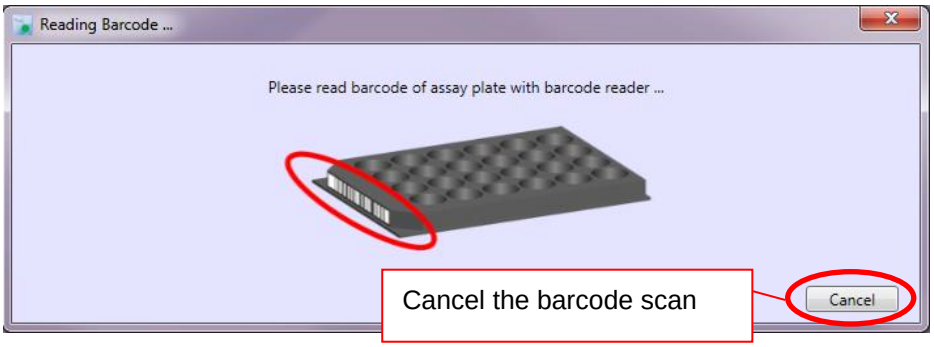
8) Enter appropriate text in the “Source Plate” field for source plate name.



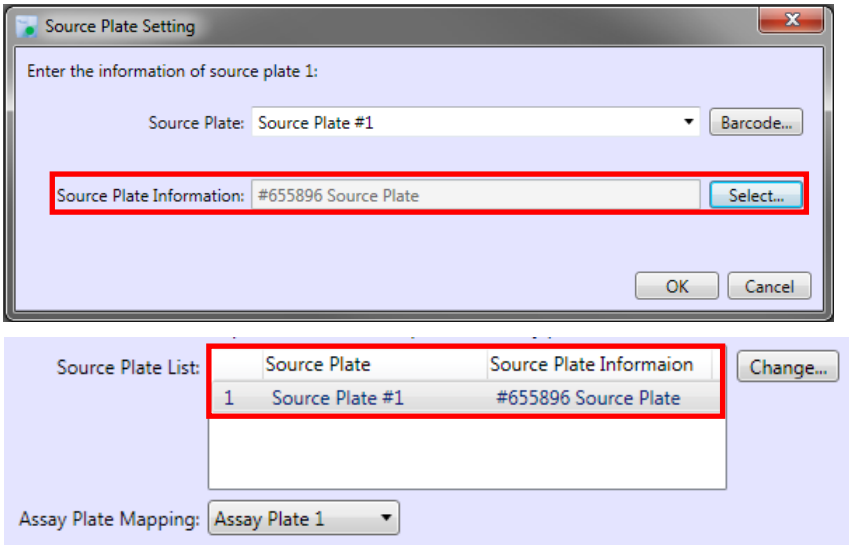
Click “Barcode” If there is a barcode on the Source plate.
(Barcode reader model only)



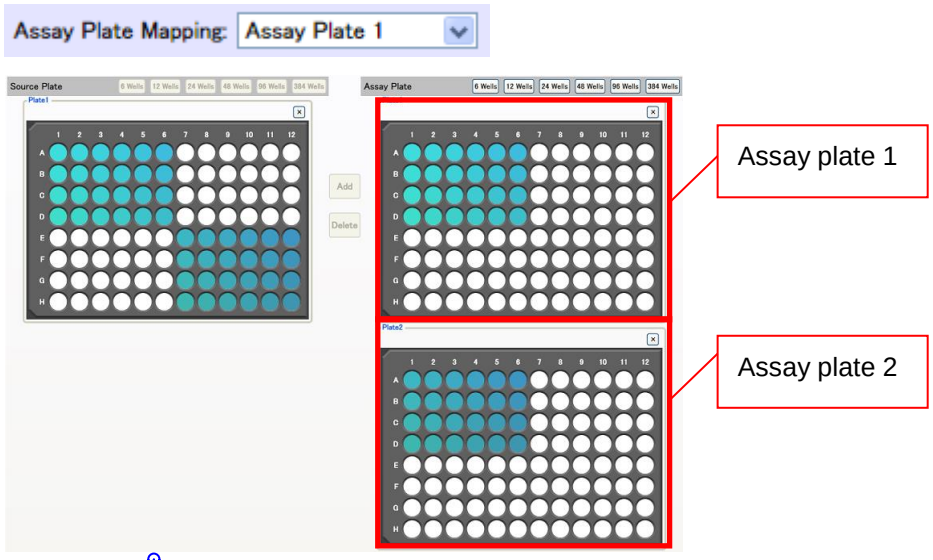
Barcode reader starts to scan. The scanned barcode data is entered to the “Source Plate” field.



9) Click “Select” and select a well plate information file for the source plate. (Refer to 4.1)



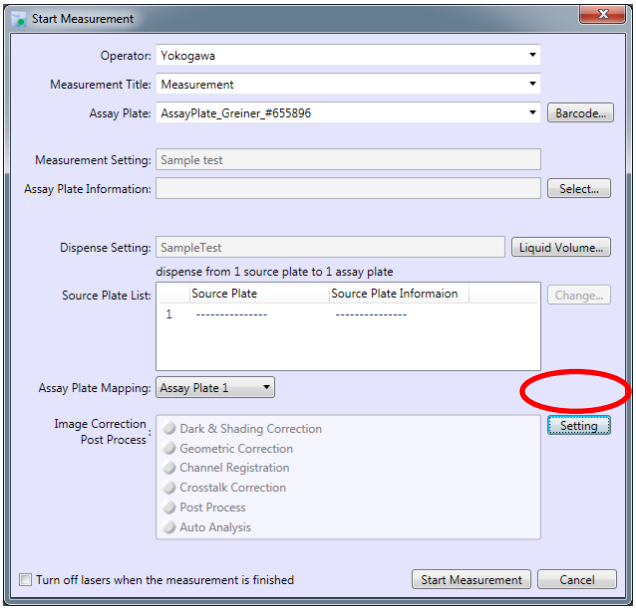
10) Select “Assay Plate Mapping.” If multiple assay plates were set in the dispensing setting file (refer to 5.12), solution is dispensed only to the wells in the assay plate selected in “Assay Plate Mapping.”



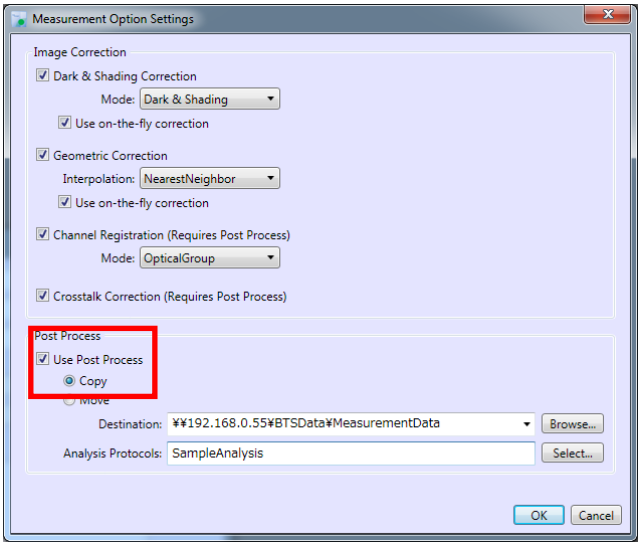
MEMO

●Multiple assay plates cannot be measured.

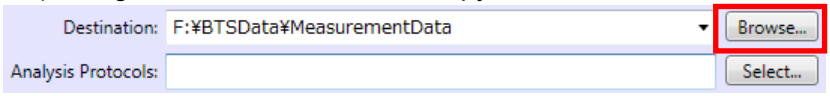
11) Click “Setting” if using post-processing function.



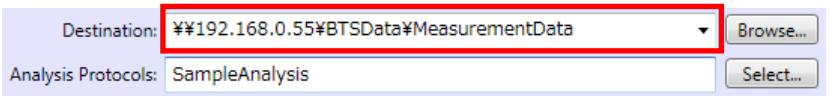
12) “Measurement Option Settings” window opens.
In case of using post-processing function, check “Use Post Process”.
Select “Copy” to automatically copy the measurement data to the designated folder. Select “Move” to automatically move the measurement data to the designated folder.



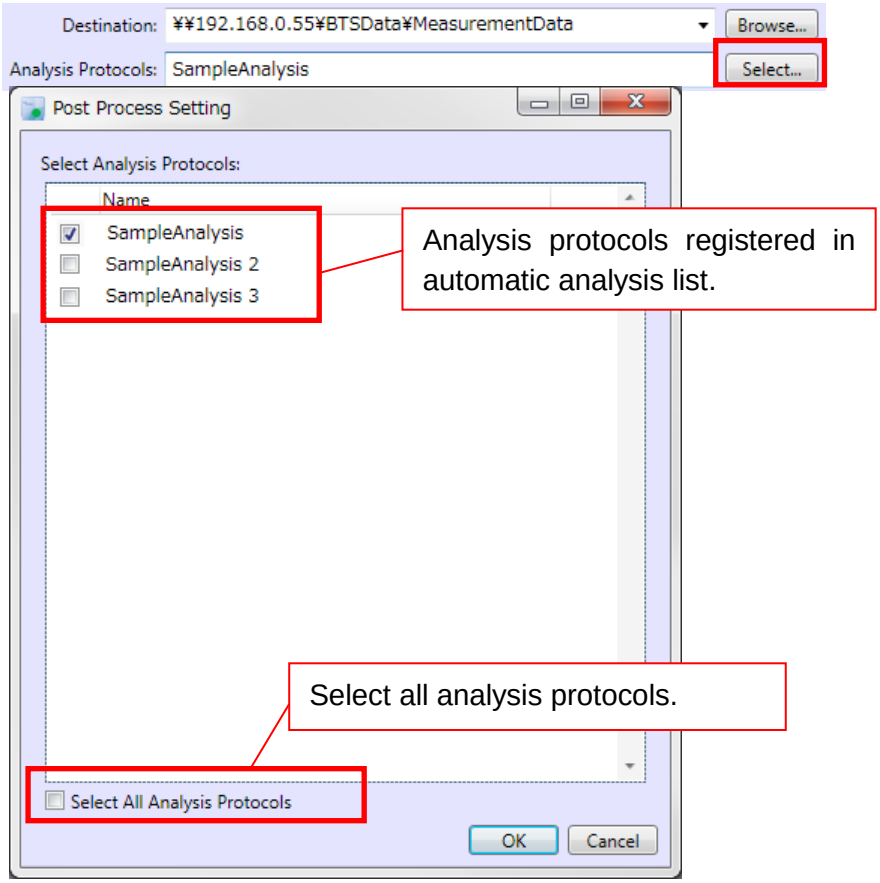
13) Designate the folder where to copy or move data from “Select”.



To perform automatic analysis, specify a folder selectable from CV7000 Analysis Software.
(Example: Specify “D:¥BTSData¥MeasurementData” in the WS for analysis as “Destination.”)



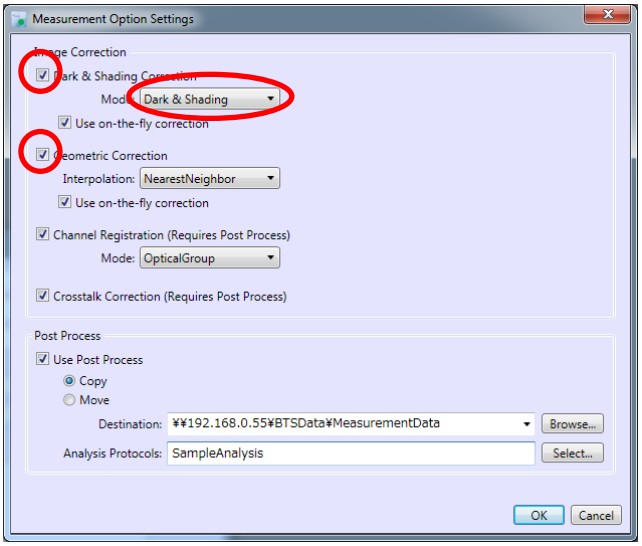
To perform automatic analysis, click “Select” of “Analysis Protocol” and then, select analysis protocols to be analyzed.
(About analysis protocol, refer to section 12.6 of IM 80H01C03-01E CV7000 Analysis Software User’s Manual)
Click “OK” after selecting.



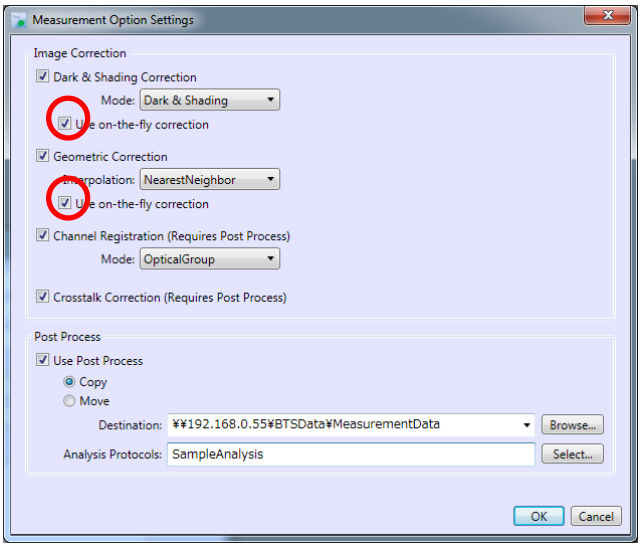
MEMO

●To display the AnalysisProtocol setting in the Start Measurement screen, launch the CV7000 Analysis Software and set to communicate the network previously.

14) Set image correction setting post imaging (for detail of image correction, please refer to IM 80H01A16-01E Image Correction Software User's Manual). Set "Dark & Shading Correction" and "Geometric Correction". "Dark & Shading" is recommended for "Dark & Shading Correction".

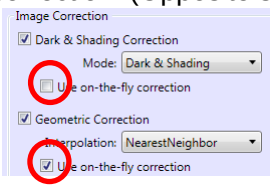


15) Check if performing on-the-fly correction.

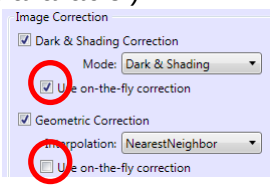


MEMO

- If "Use on-the-fly correction" is checked, image correction is performed before saving image to hard disk.
  Processing speed becomes faster than using Post Processor, but raw image data is not saved.
- It cannot to perform "Dark & Shading Correction" without using on-the-fly correction and perform "Geometric Correction" with using on-the-fly correction. (Opposite setting is available.)

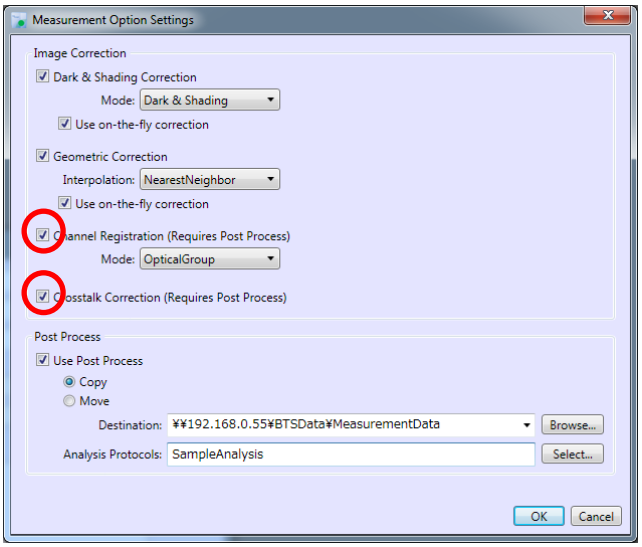


← NG



← OK

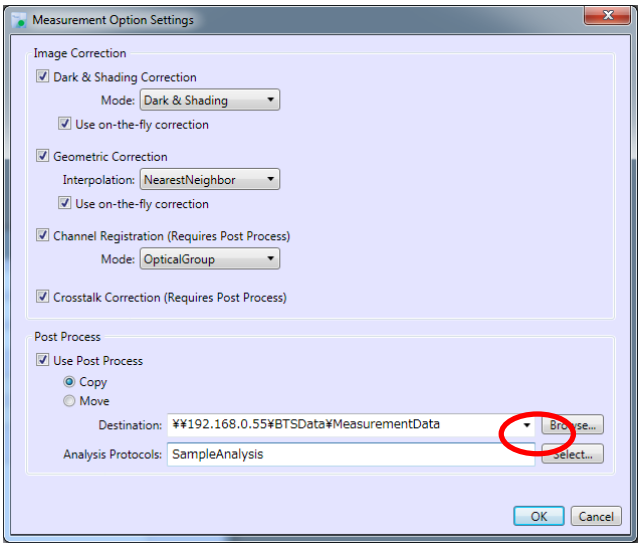
16) Set “Channel Registration” and “Crosstalk Correction”. These 2 items are selectable if “Use Post Process” is checked. Also, they are not correspond to on-the-fly correction.



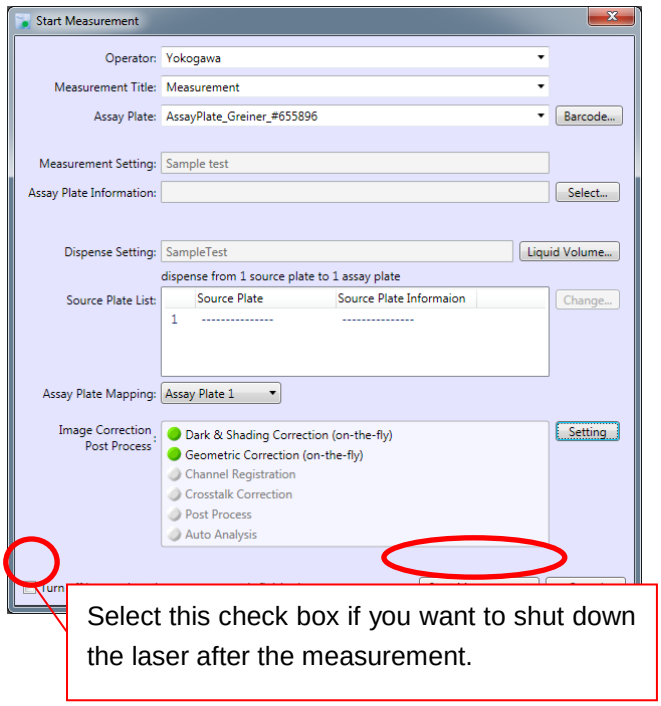
MEMO

- ●Don‘t select “Channel Registration” if the images are acquired by high-speed time-lapse mode.
- ●To correct fluorescent crosstalk, “Dark & Shading” must be selected in “Dark & Shading Correction”, and “Geometric Correction” or “Channel Registration” must be selected.
  It is recommended to select both “Geometric Correction” and “Channel Registration”.
- ●”Crosstalk Correction” cannot be performed if “Fluorophore” is not designated in Channel Setting (refer to 5.5)
- ●Emission spectrum of fluorophore can be deformed by surrounding environment (pH, temperature, saline concentration). In this case, crosstalk correction cannot be performed normally.
  Confirm image after crosstalk correction by Image Correction Software and if the images have abnormality, uncheck “Crosstalk Correction”.

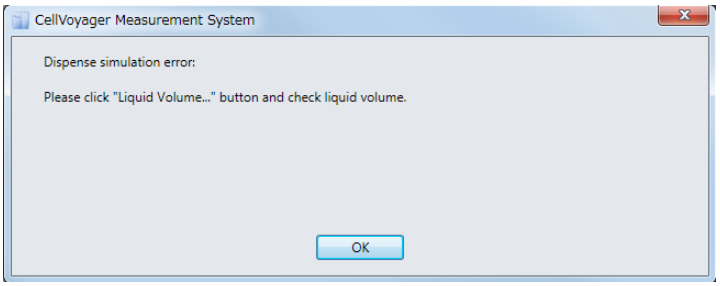
17) Click “OK”.



18) Click “Start Measurement”.

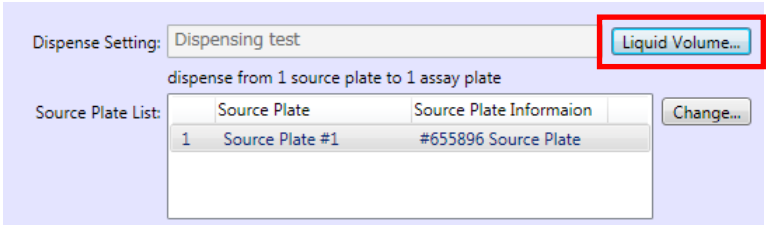


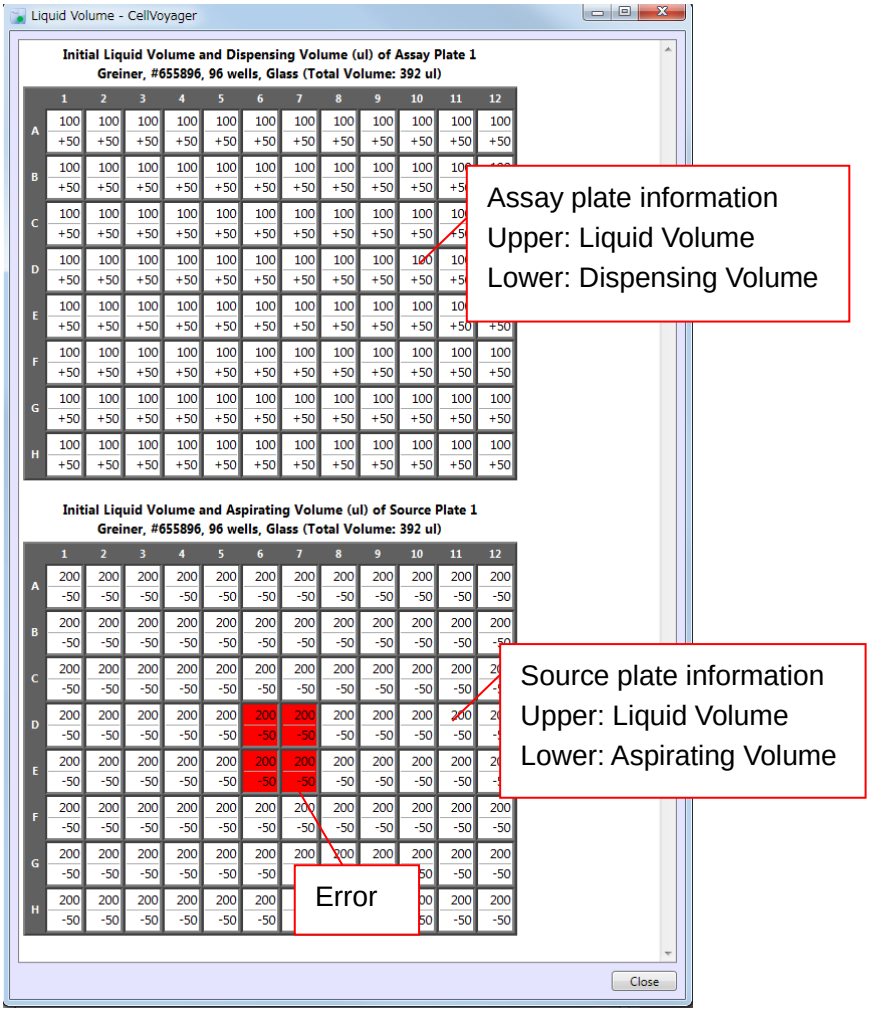
Measurement cannot start if any error is contained in the result of simulation in dispensing setting screen. (Refer to 5.12.)



MEMO

●In the case of error, click “Liquid Volume” to confirm error point. To check error in detail, confirm result obtained by dispensing simulation in dispensing setting screen. (Refer to 5.12.)





Liquid Volume screen

19) Set well plates on the sample loader. (Refer to 8.1)
Measurement will start once the plates are set.

⚠ WARNING

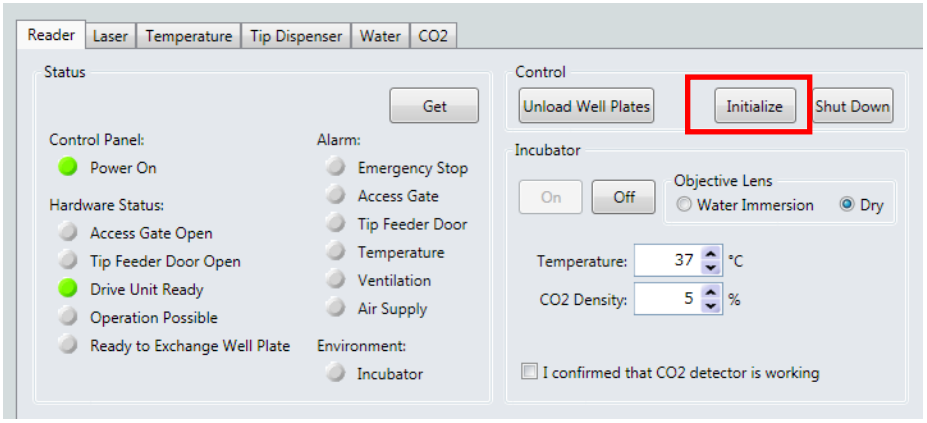
- In the case of dispensing measurement, be sure to remove the covers of the assay plate and source plate before setting the plates. Failure to do so may cause dispenser damage.
- The Assay plate and Source plate simulated in the dispensing setting file (refer to 5.12) must be used in dispensing measurement. Failure to do so may cause dispenser damage.

20) When the measurement is complete, remove the well plates. (Refer to 8.2)

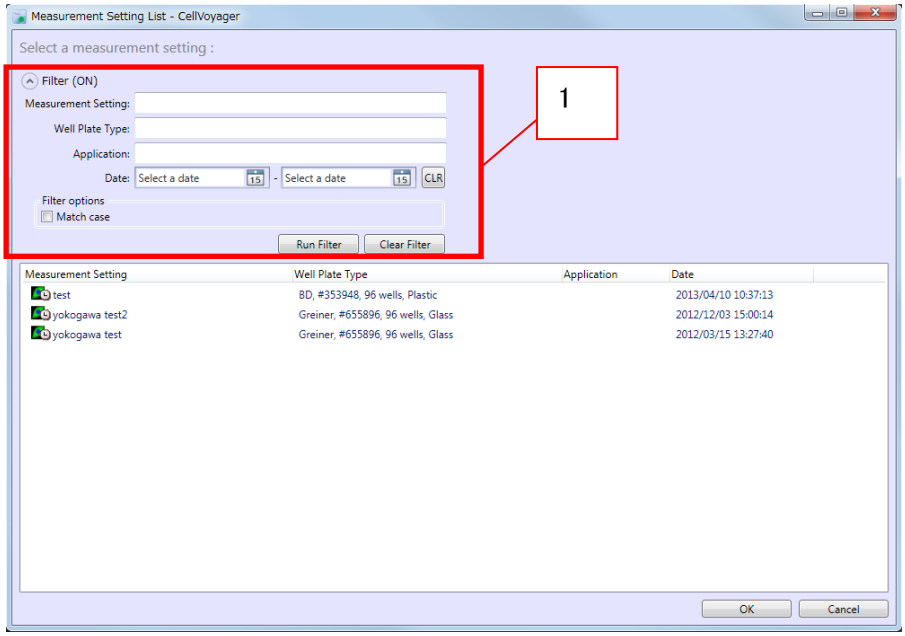
21) Measured results are displayed. (Refer to Chapter 9)

MEMO

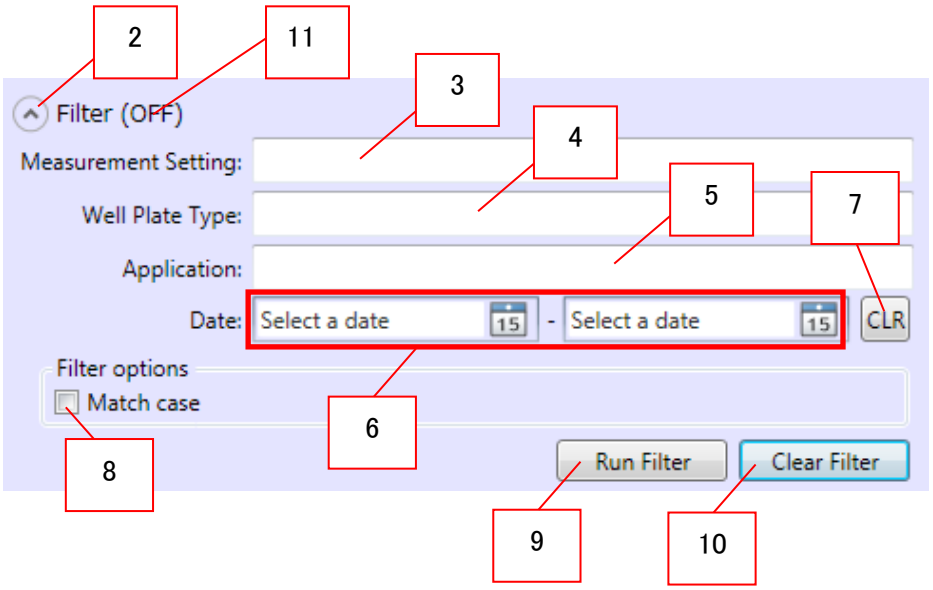
- To start the laser again, click “Initialize” on the Reader Status tab of the Reader Control screen.



8.5. Measurement Setting List Screen

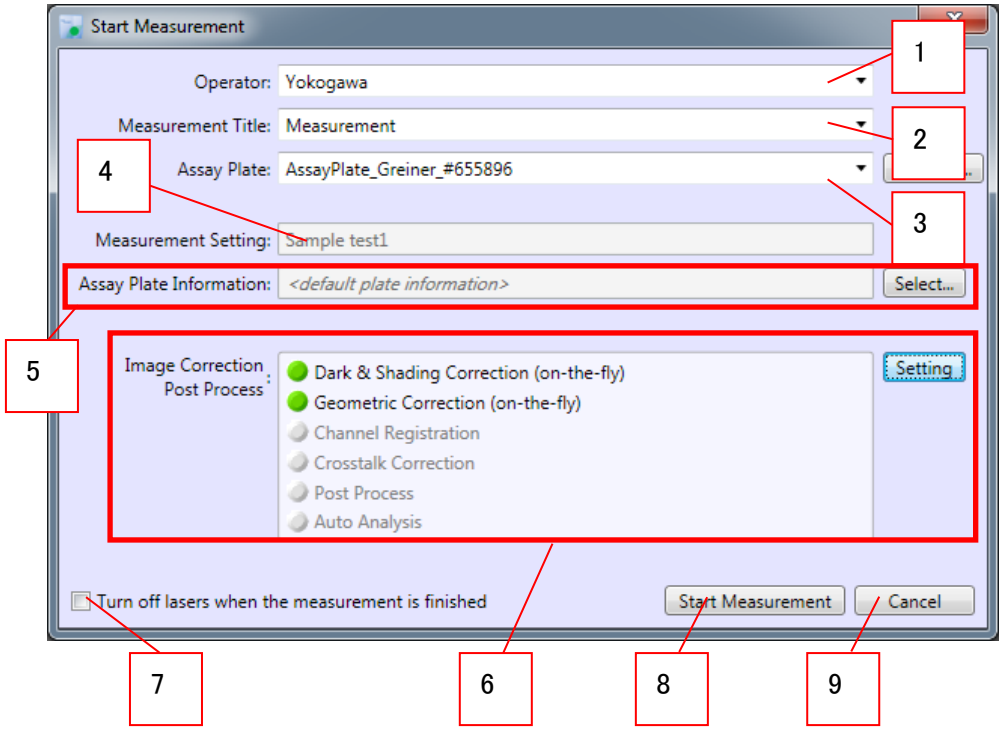


1) Screen for search filter setting

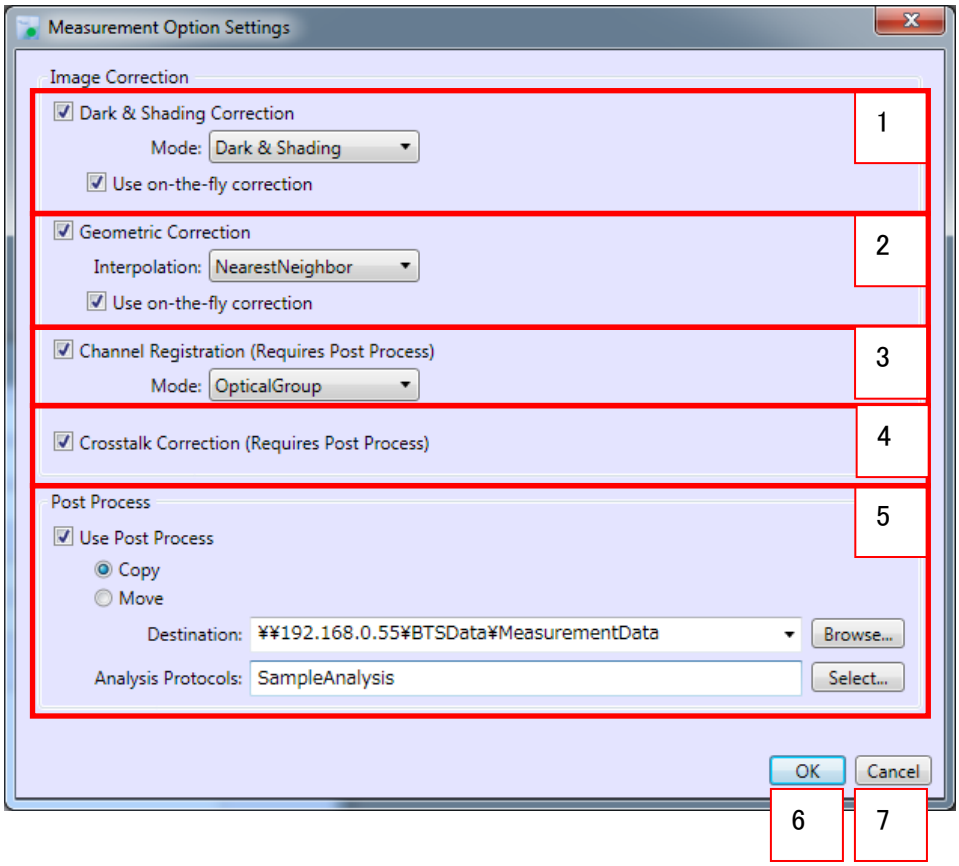


2) Display the screen for search filter setting.
3) Character string of Measurement Setting to perform search filter.
4) Character string of Well Plate Type to perform search filter.
5) Character string of Application to perform search filter.
6) Date range to perform search filter.
7) Clear Date range you have specified.
8) Perform search filter by case-sensitive.
9) Perform search filter.
10) Clear result of search filter.
11) Search filter ON/OFF

8.6. Start Measurement Screen



1) Operator name
2) Measurement title
3) Assay plate name
4) Measurement setting file
5) Well plate information file of the Assay plate
6) Setting items of postprocessor and image correction.
   (Click “Setting” button to open setting screen described in next page.)
7) Automatically turns OFF laser power when the measurement finished.
8) Start measurement.
9) Cancel measurement.



1) Background/ Shading correction setting

Item		Explanation
Dark & Shading Correction		Correct the background offset level and shading of measured images.
Mode	Dark & Shading	Correct the background offset level and shading of measured images.
	Dark	Correct the background offset level of measured images.
Use on-the-fly correction		Perform on-the-fly correction

2) Geometric correction setting

Item		Explanation
Geometric Correction		Correct scaling, rotation and parallel shift effects.
Interpolation	NearestNeighbor	Use the value of the nearest pixel to correct images.
	Bilinear	Decide the pixel value by the distance and variation of each of nearby four pixels.
Use on-the-fly correction		Perform on-the-fly correction

3) Registration correction setting

Item		Explanation
Channel Registration		Correct pixel shift between channels.
Mode	CompareBased	Correct pixel shift by comparing the correlation between the reference channel and each channel.
	OpticalGroup	Correct pixel shift by comparing the correlation between groups of each XY stage position.

4) Setting of crosstalk correction

5) Setting items of postprocess

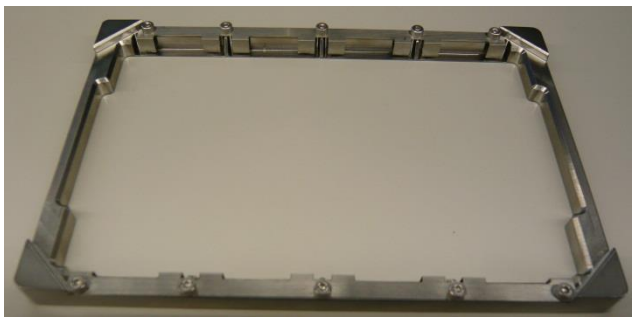
Item	Description
Use Post Process	Post processing starts after measurement completed when checked.
Copy	Automatically copy measurement data to designated folder.
Move	Automatically move measurement data to designated folder.
Destination	Designate folder to copy or move data by “Select”. To perform automatic analysis, specify a folder selectable from the Analysis Support software.
Analysis Protocol	Specify analysis protocols by clicking “Select.” (Analysis software supported model only)

6) Close the screen with saving image correction/ post process setting.

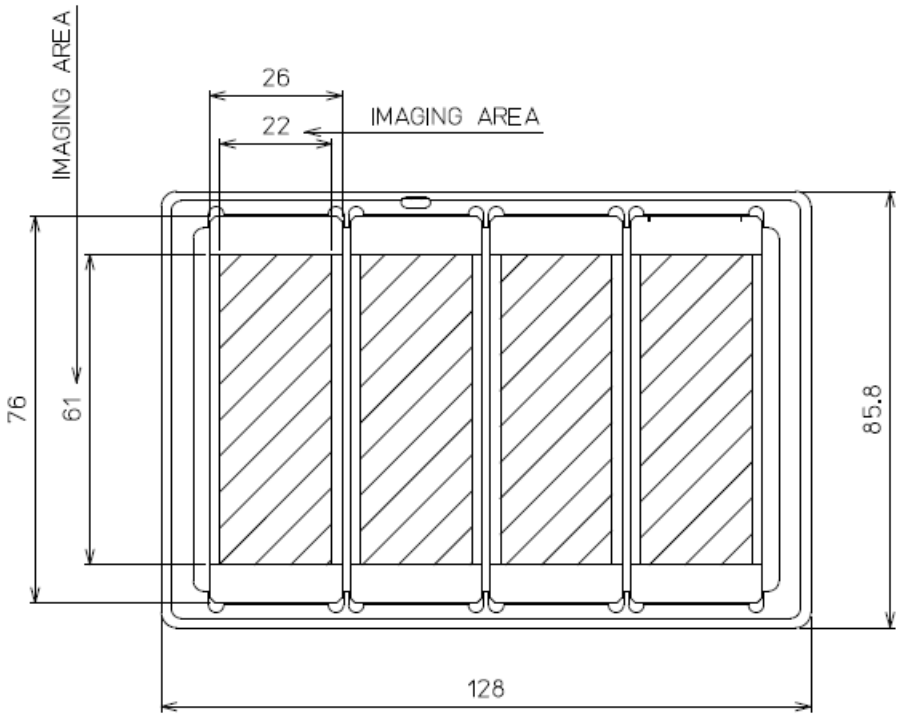
7) Close the screen without saving image correction/ post process setting.

8.7. Set Slide Glasses on Slide Glass Holder

In the case of observation by slide glass, set slides on slide glass holder. (Only slide glass holder supported model.)

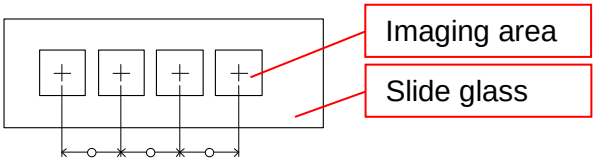


Slide glass holder

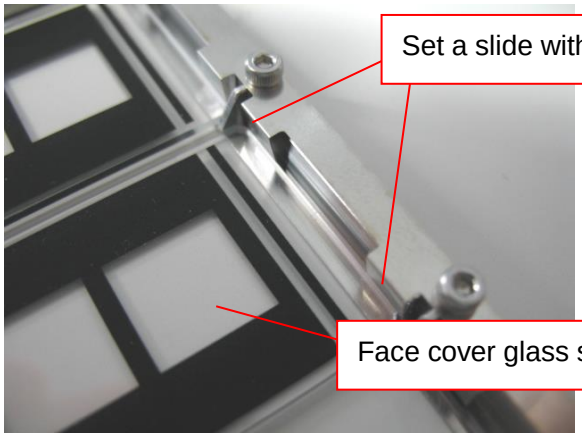


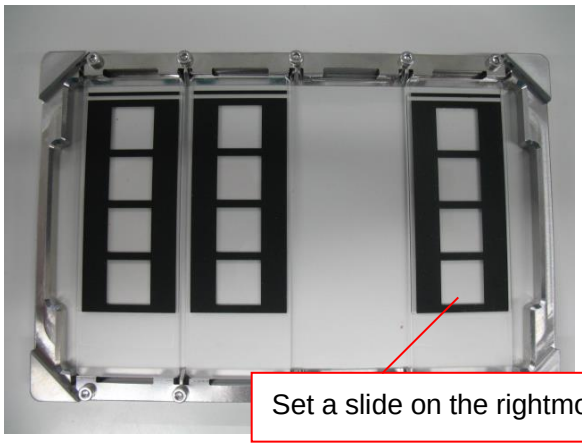
MEMO

- Up to 4 slide glasses can be set.
- Observable range of slide glass is 61mm X 22mm.
  The range can be small when water immersion lens is used.
- Slide glass that can be analyzed by CV7000 Analysis Software is what 4 imaging areas are arranged at regular intervals as following diagram.



1) Set a slide on sample holder with pressing to leaf spring. Face cover glass side down. Please be careful not to tilt slides.





⚠ WARNING

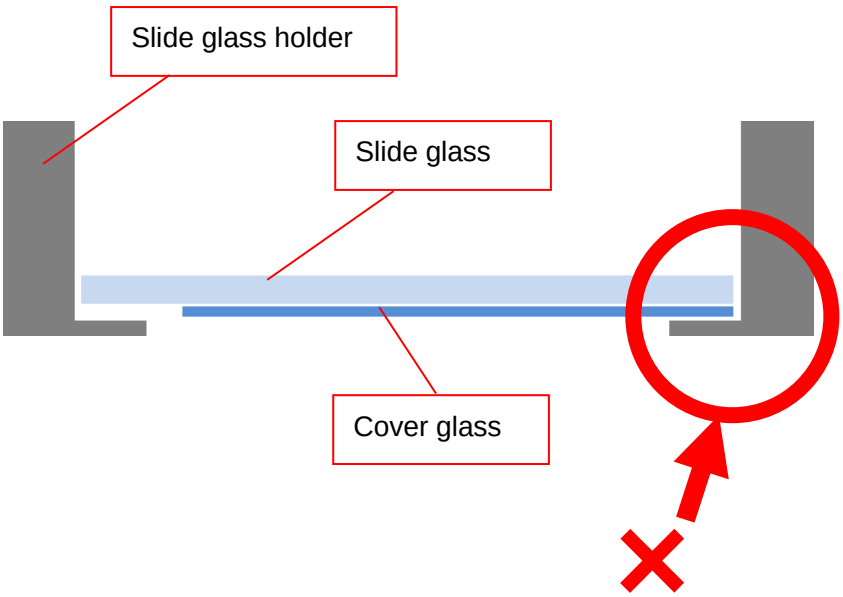
- Supported silde glasses are as following which meet ISO 8073/1 (JIS R 3703).
  Length [mm]: 76.0 0/-1
  Width [mm]: 26.0 0/-1
  Don't use slide glasses not to meet above condition. Slide glasses could be broken if they are set by sheer force. Besides, slide glasses could fall into equipment during measurement and injure the equipment.
- Cover glass must be fixed on slide glass by manicure and others not to fall into machine.

⚠ CAUTION

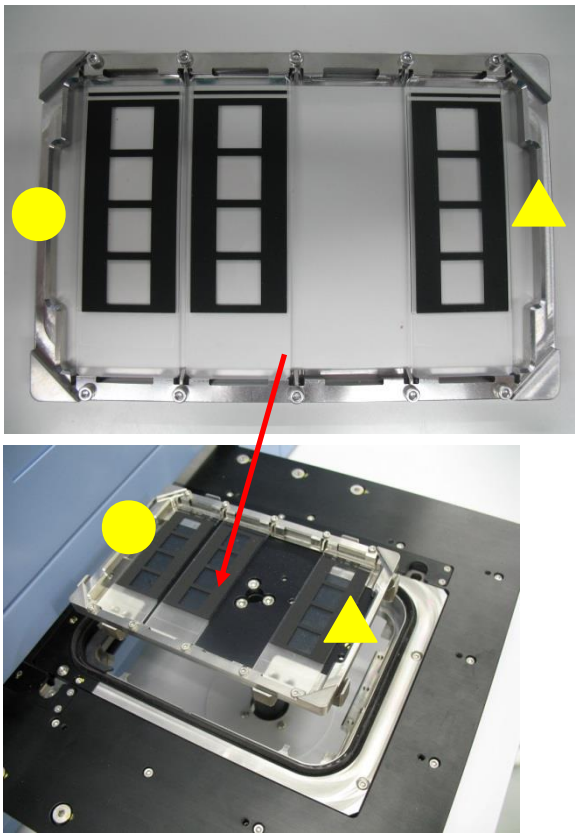
- If the slide glasses not to meet ISO/ JIS, slide glass can't be fixed on slide glass holder and imaging area shift or focus error could occur.
- Set a slide glass on the rightmost position of holder. Sample isn't needed to set on this slide glass.

⚠ CAUTION

●Set slides not to put cover glass on sample holder.



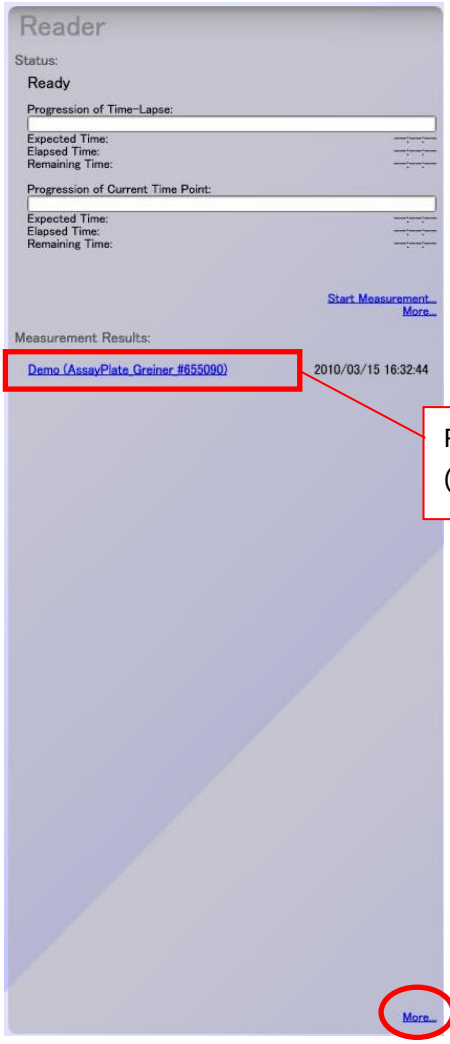
3) Set slide glass holder on the stage. Be careful that direction of holder is correct. For the procedure of setting to the stage, please refer to 8.1.



9. Checking Measured Images

9.1. Opening the Measurement Result Screen

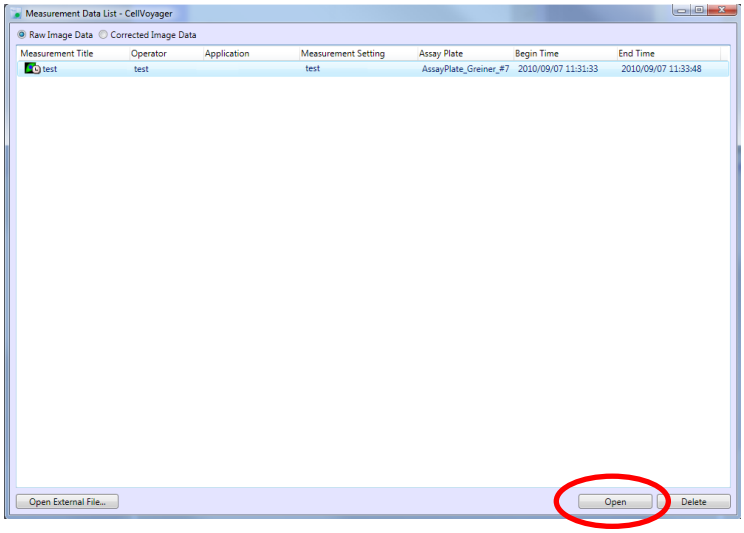
1) Click “More” under “Measurement Results” in the Reader Area.



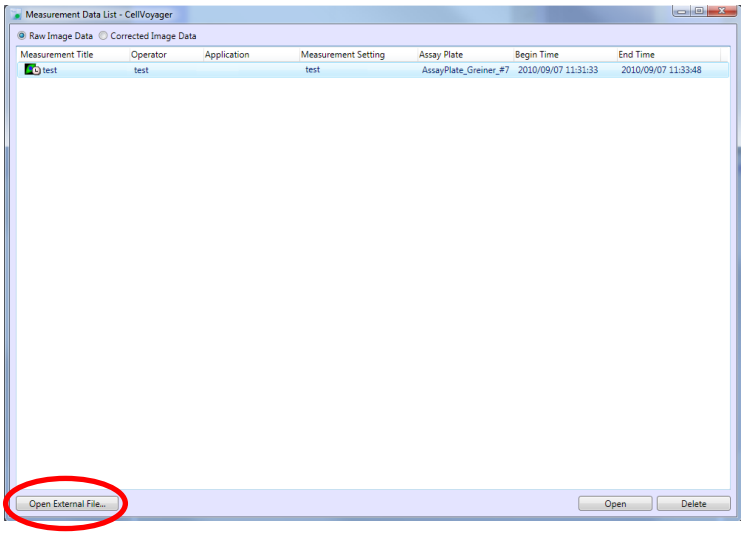
Recently measured or referenced data
(Clicking here displays measured data.)

Browse measured data

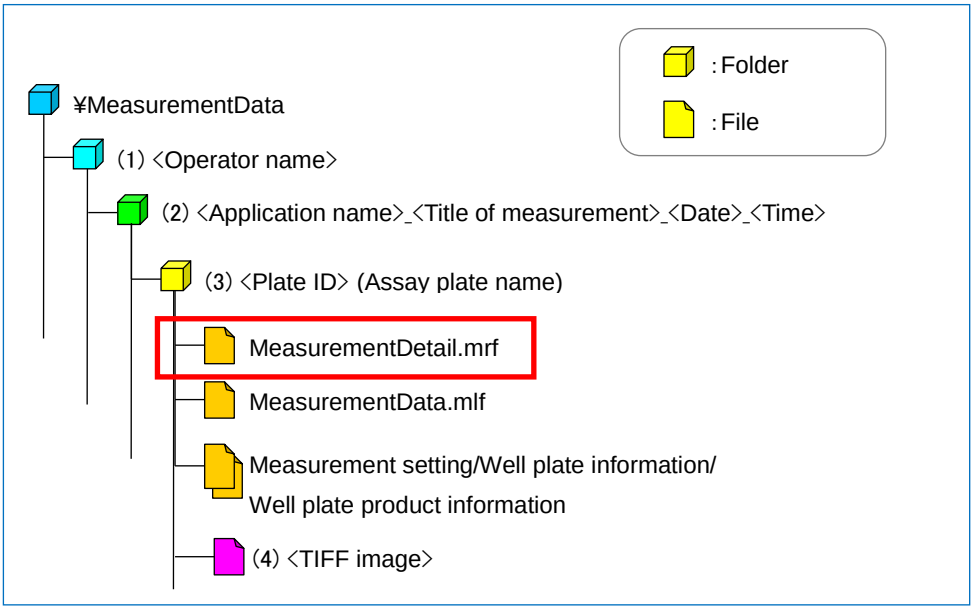
2) The Measurement Data List screen opens. (Refer to 9.3)
Select a desired measurement data and then click “Open.” The measured result is shown.

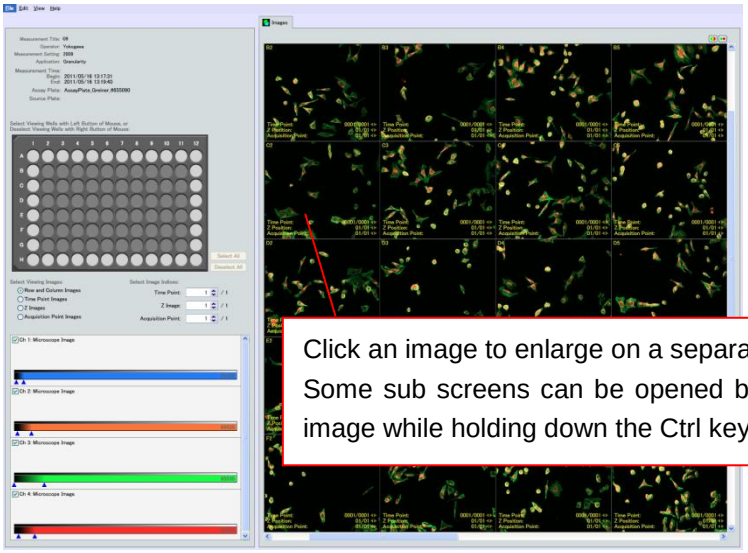


Click “Open External File” if you open the measured result from the external folder you saved.

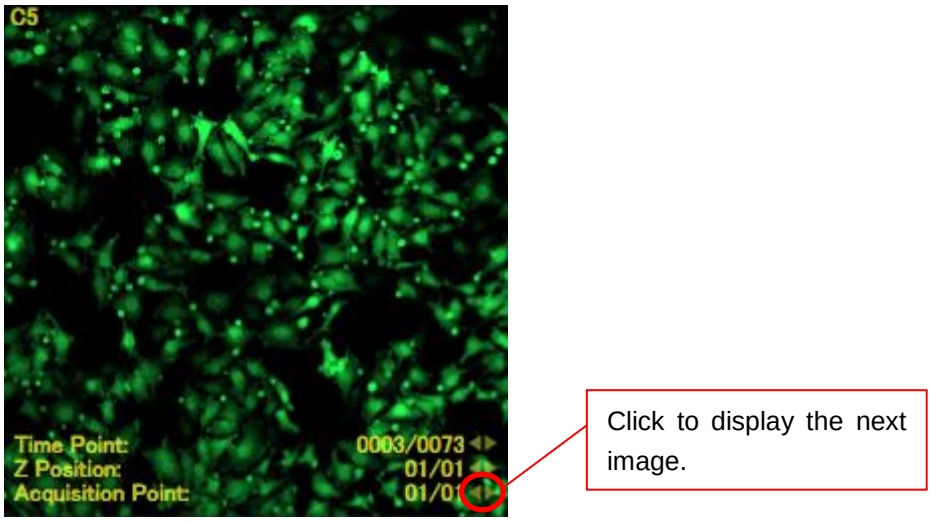


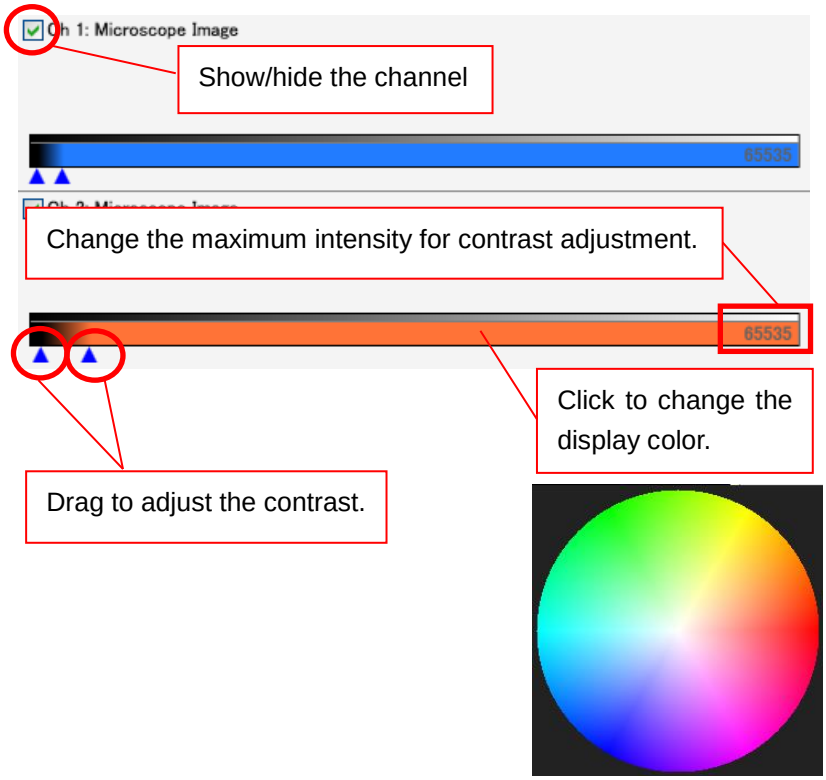
Select the “MeasurementDetail.mrf” file in the measurement folder. The measured result is shown.

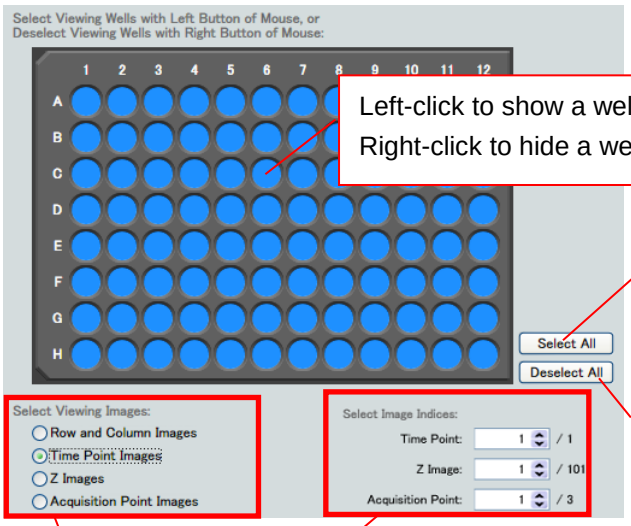




Output screen for measured results (Refer to 9.3)







Left-click to show a well.
Right-click to hide a well.

Show all wells.

Hide all wells.

Change the number of all images displayed on the display area.

Row and column Images：Show images in the well plate view.
Time Point Images：Show images along the time axis.
Z Images：Show images for each Z slice.
Acquisition Point Images：Show images for each acquisition point.

Change the well to display.

Change the Z position to display.

Snapshot

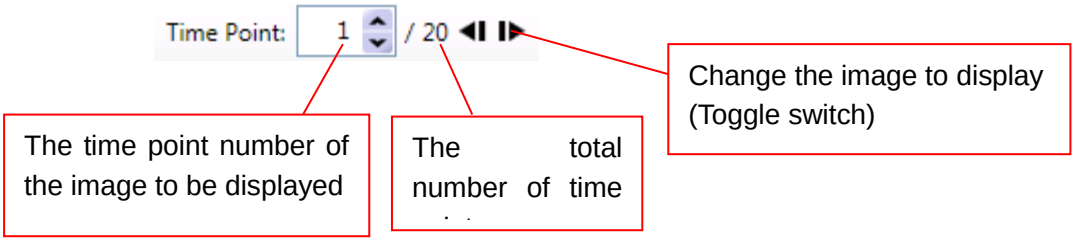
Change the time point to display.

Change the field to display.

Scroll the mouse wheel to zoom into/out of the image.

Pixel coordinate and each channel of intensity

Sub screen for measurement results



“Z Position” and “Acquisition Point” is in common with “Time Point”

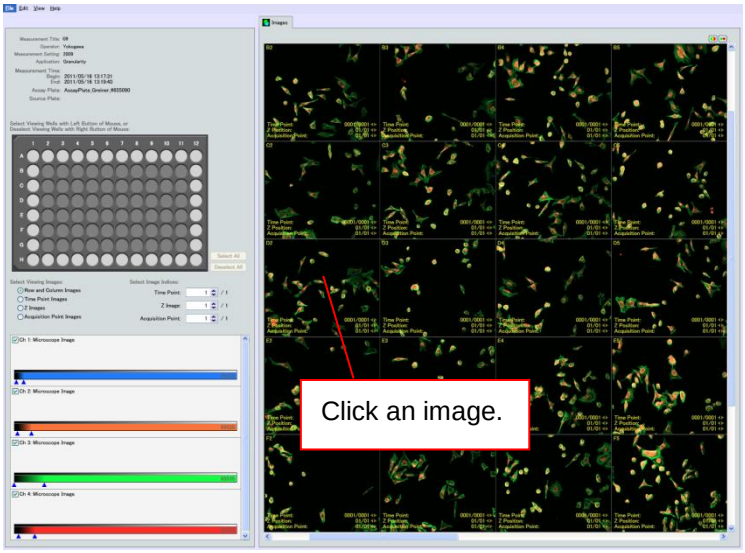
9.2. Creating Movie Files

Movie files can be created from the measurement data acquired by time-lapse imaging.

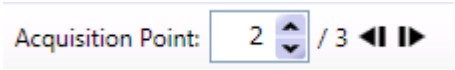
Creating the Movie File for a Single Field

The movie file about the field displaying on the sub screen is created.

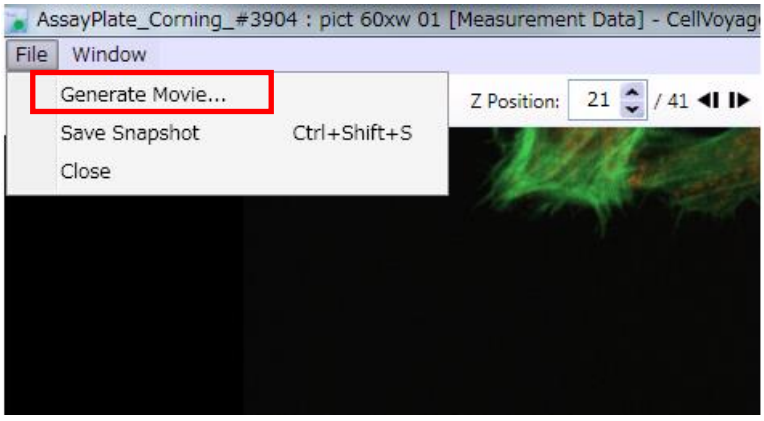
1) An image is displayed to the sub screen by clicking a well image to create movie files. (Refer to 9.3.)



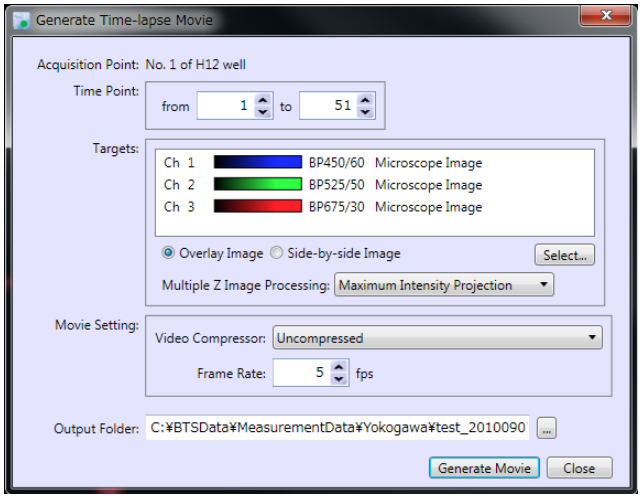
2) From the sub screen, select the field to create movie files.



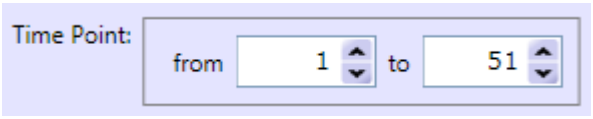
3) Click “Generate Movie” from the “File” menu of the sub screen.



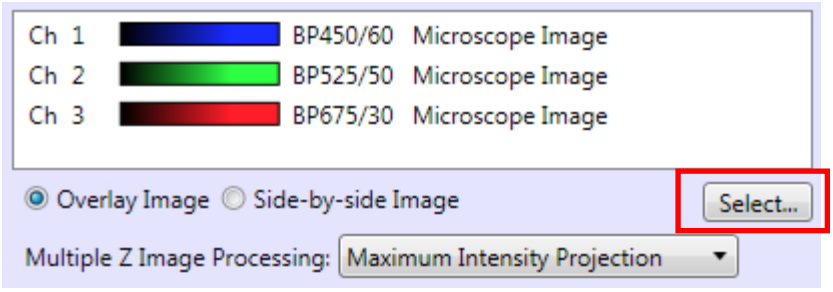
The “Generate Time-lapse Movie” screen is displayed.

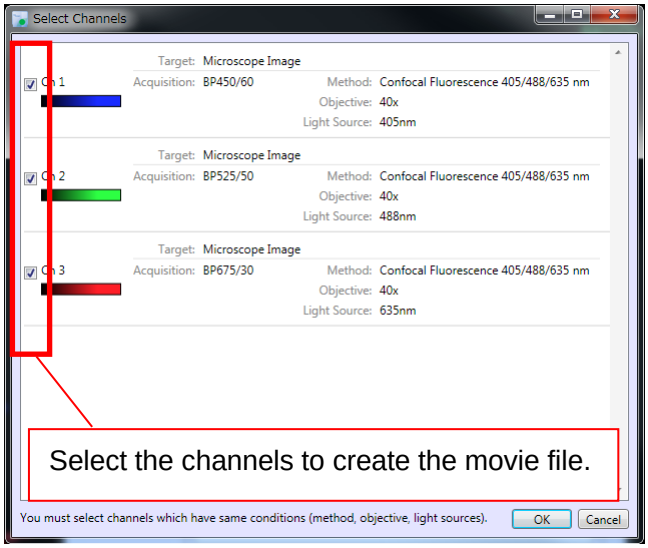


4) Specify the time point range to create movie files.

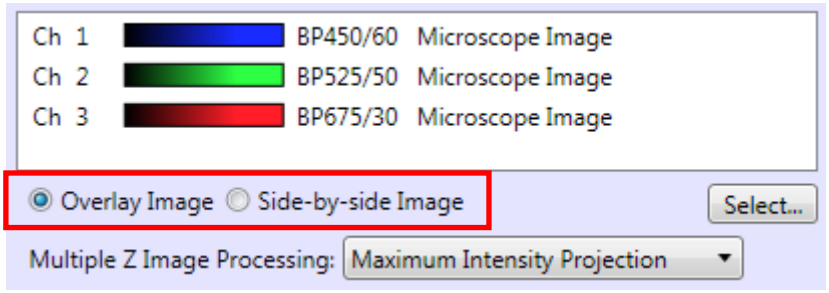


5) Select the channels to create the movie file by clicking “Select.”

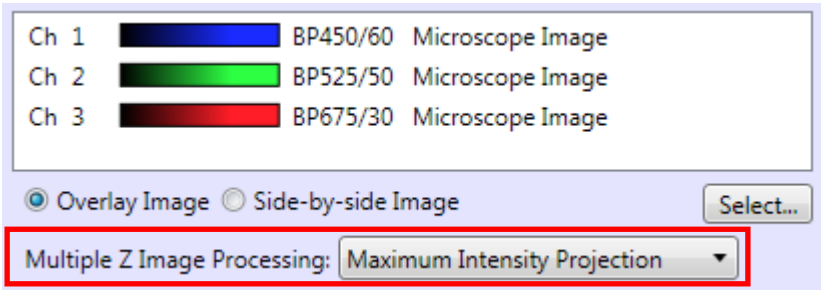




6) If selecting “Overlay Image,” the movie file is created with overlaying channel images.
If selecting “Side-by-side Image,” the movie file is created with tiling channel images.

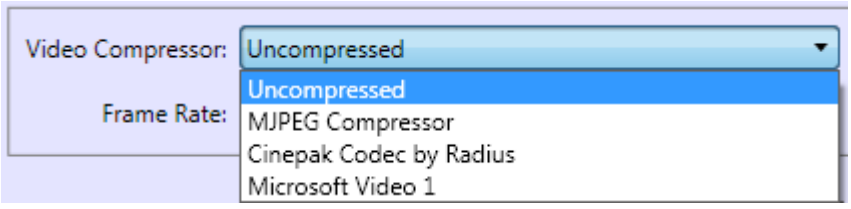


7) Select an output item for Z images from “Multiple Z Image Processing,” if selecting the channels which have Z-stack images.

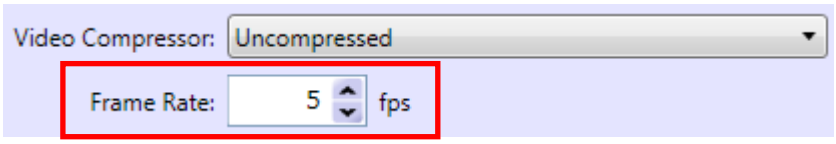


Item	Explanation
None	No Z-stack image exists.
Maximum Intensity Projection	Create movie files using the MIP images. (Refer to 5.1.)
Minimum Intensity Projection	Create movie files using the MinIP images. (Refer to 5.1.)
Average Intensity Projection	Create movie files using the AIP images. (Refer to 5.1.)
Maximum Intensity Image	Create movie files using the Z images which have the highest total intensity in the Z-stack images at each time point.
Sum Intensity Projection	Create movie files using SUM images. (Refer to 5.1.)

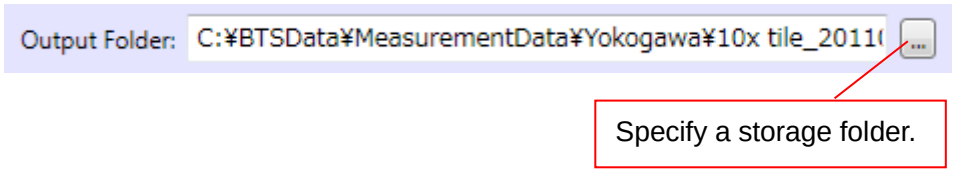
8) Select a compression method for movie files from “Video Compressor.”
If compressing movie files, “Cinepak Codec” is recommended.
If not compressing movie files, select “Uncompressed.”



9) Specify the frame rate for movie replay.



10) Specify a storage folder for movie files.



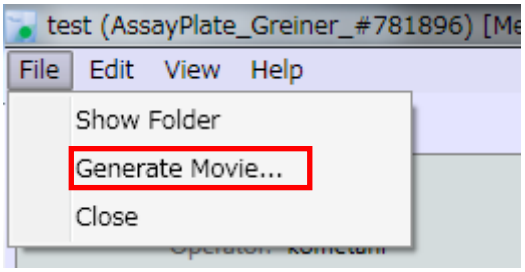
11) To create movie files is started by clicking “Generate Movie.”

The screen to display the processing for creating movie files

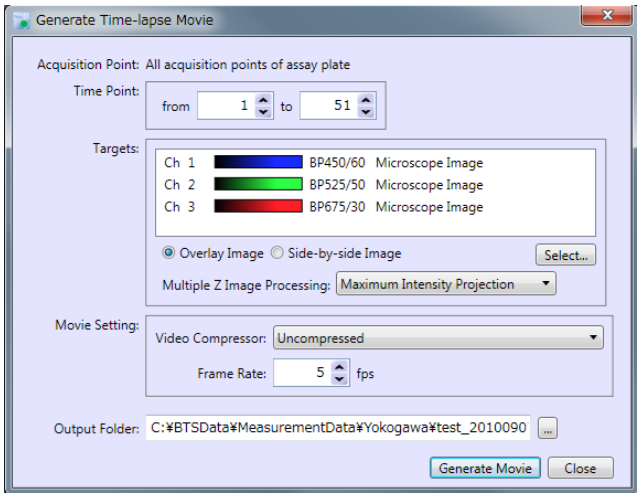
Creating the Movie Files for All Fields

The movie files for the entire fields in all wells are created.

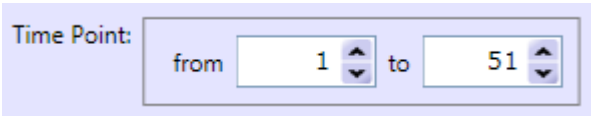
1) Click “Generate Movie” from the “File” menu in the measurement result screen. (Refer to 9.3.)



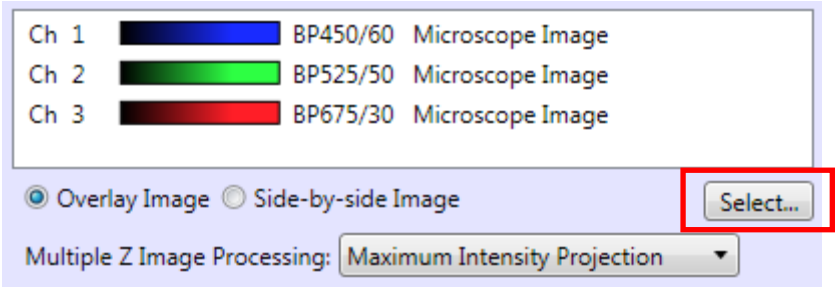
The “Generate Time-lapse Movie” screen is displayed.

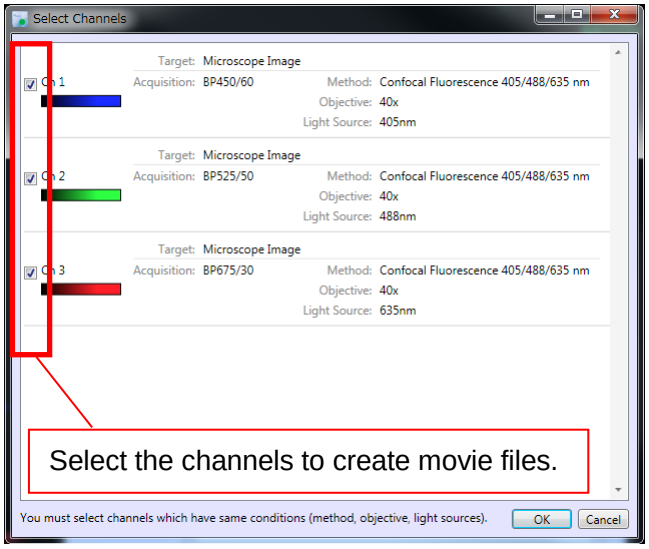


2) Specify the time point range to create movie files.

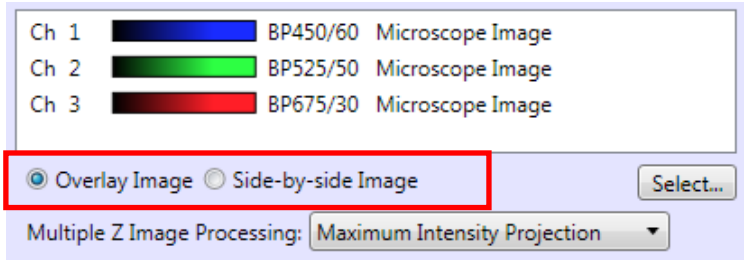


3) Select the channels to create movie files by clicking “Select.”

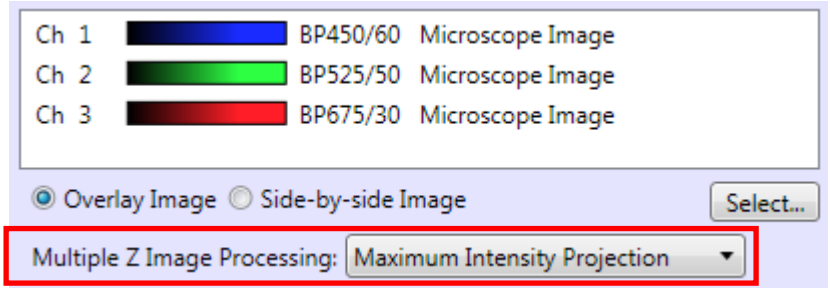




4) If selecting “Overlay Image,” movie files are created with overlaying channel images.
If selecting “Side-by-side Image,” movie files are created with tiling channel images.

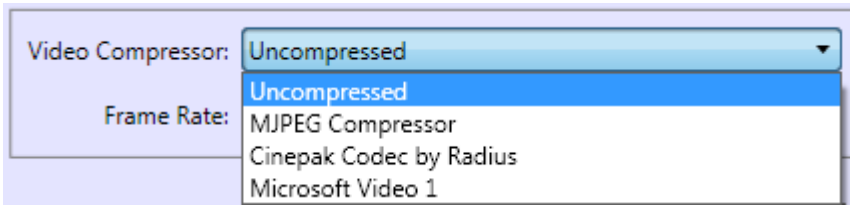


5) Select an output item for Z images from “Multiple Z Image Processing,” if selecting the channels which have Z-stack images.

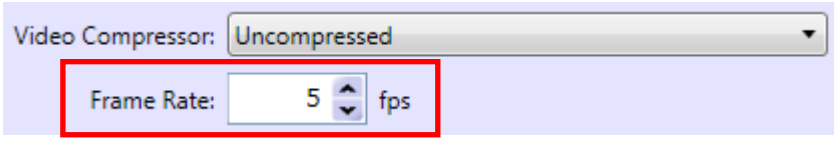


Item	Explanation
None	No Z-stack image exists.
Maximum Intensity Projection	Create movie files using the MIP images. (Refer to 5.1.)
Minimum Intensity Projection	Create movie files using the MinIP images. (Refer to 5.1.)
Average Intensity Projection	Create movie files using the AIP images. (Refer to 5.1.)
Maximum Intensity Image	Create movie files using the Z images which have the highest total intensity in the Z-stack images at each time point.
Sum Intensity Projection	Create movie files using SUM images. (Refer to 5.1.)

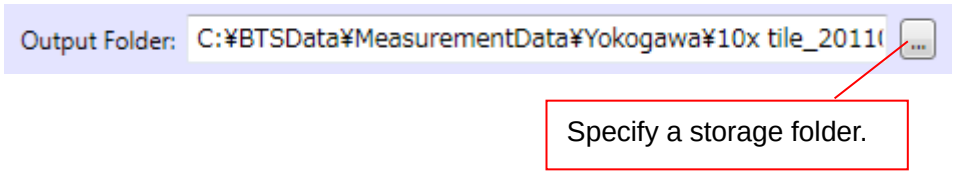
6) Select a compression method for movie files from “Video Compressor.”
If compressing movie files, “Cinepak Codec” is recommended.
If not compressing movie files, select “Uncompressed.”



7) Specify the frame rate for movie replay.



8) Specify a storage folder for movie files.

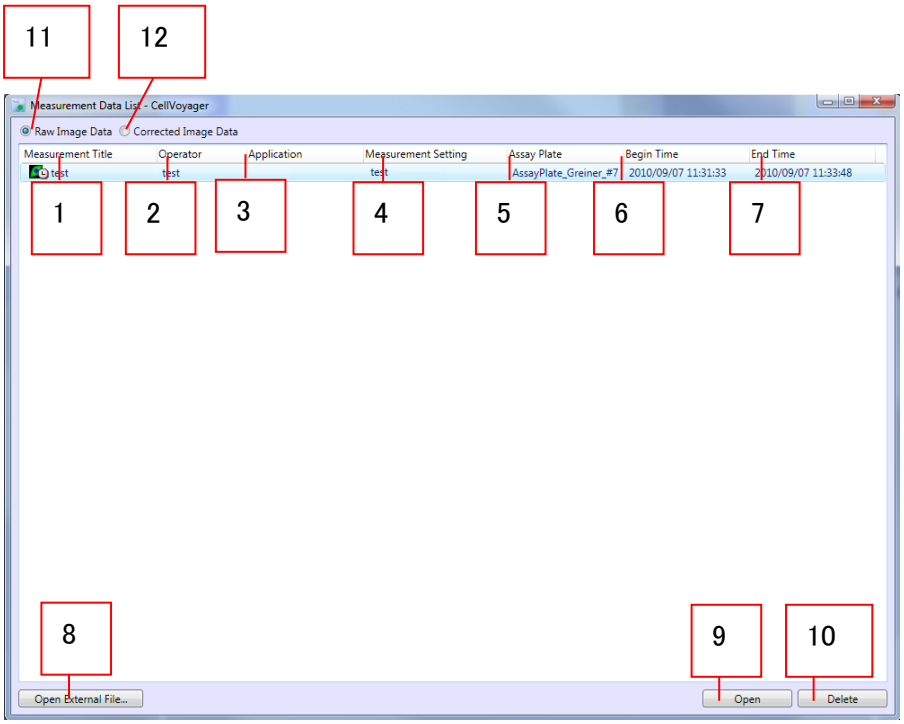


9) To create movie files is started by clicking “Generate Movie.”

The screen to display the processing for creating movie files

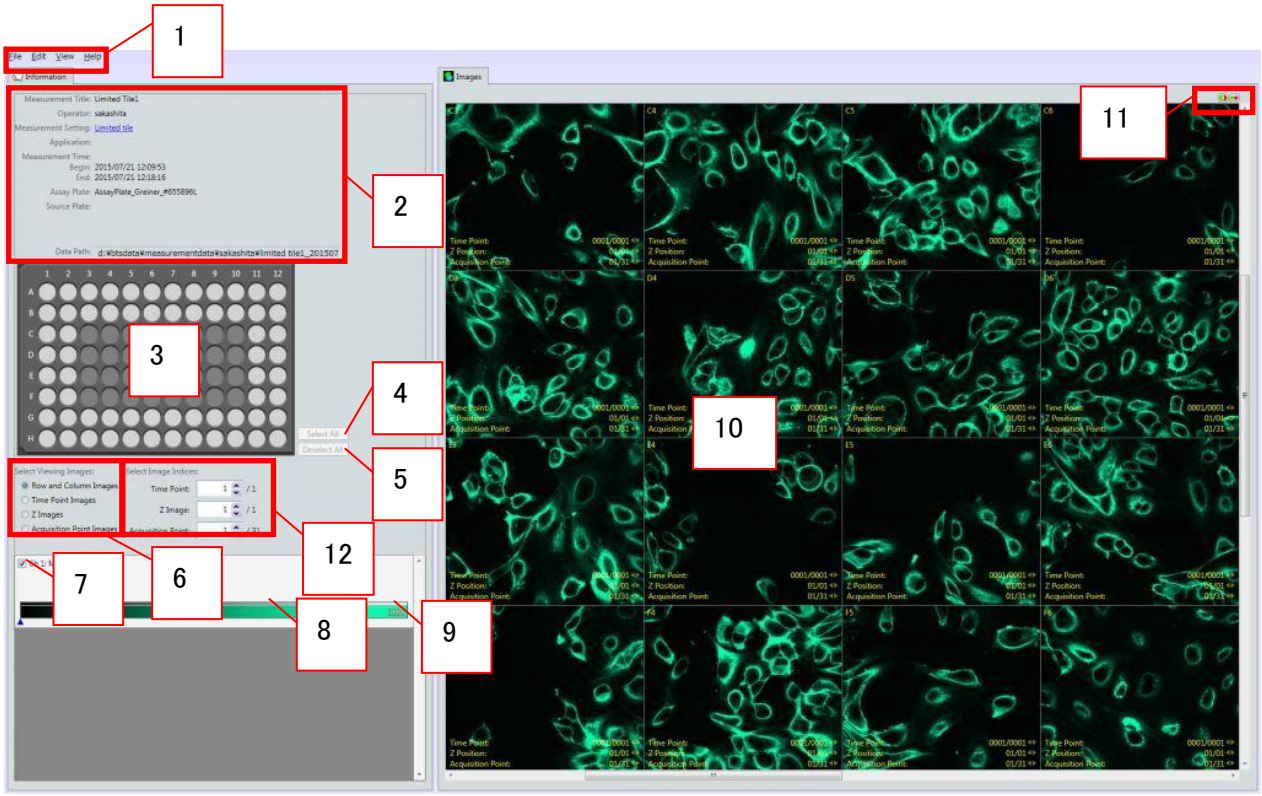
9.3. Explanation of Measurement Result Screens

Measurement Data List Screen



1) Title of measurement
2) Name of the person who performed measurement
3) Application name
4) Measurement setting file name
5) Assay plate name
6) Start time of measurement
7) End time of measurement
8) Select any measurement data.
9) Open the selected set of measured data.
10) Delete the selected set of measured data.
11) Show measured data files.
12) Show corrected image data files. (Refer to IM 80H01A16-01E)

Measurement Result Display Screen



1) Menu

File menu	Explanation
Show Folder	Display the storage folder of the measurement data.
Generate Movie	Display the screen to create movie files.
Close	Close the measurement result display screen.

Edit menu	Explanation
Undo	Undo the last operation.
Redo	Redo the last operation.
Cut	Cut the selected item.
Copy	Copy the selected item.
Paste	Paste the selected item.

View menu	Explanation
Row and Column Images	Show images in the well plate view.
Time Point Images	Show images along the time axis.
Z Images	Show images for each Z slice.
Well and Acquisition Point Images	Show images for each acquisition point.
1 Image	Show the display area by 1×1.
2 Images	Show the display area by 2×2.
3 Images	Show the display area by 3×3.
4 Images	Show the display area by 4×4.

Help menu	Explanation
About	Show the version information of the measurement software.

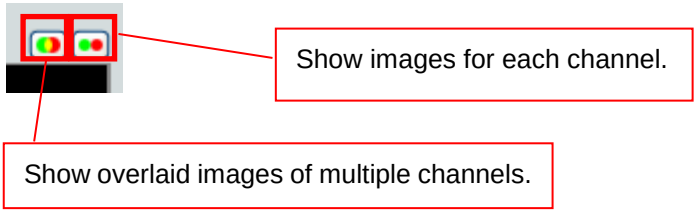
2) Information items on measured data

Item	Explanation
Measurement Title	Title of measurement
Operator	Name of the person who performed measurement
Measurement Setting	Measurement setting file name
Application	Application name
Measurement Time Begin	Start time of measurement
Measurement Time End	End time of measurement
Assay Plate	Assay plate name
Source Plate	Source plate name
Data Path	Path of data saved folder

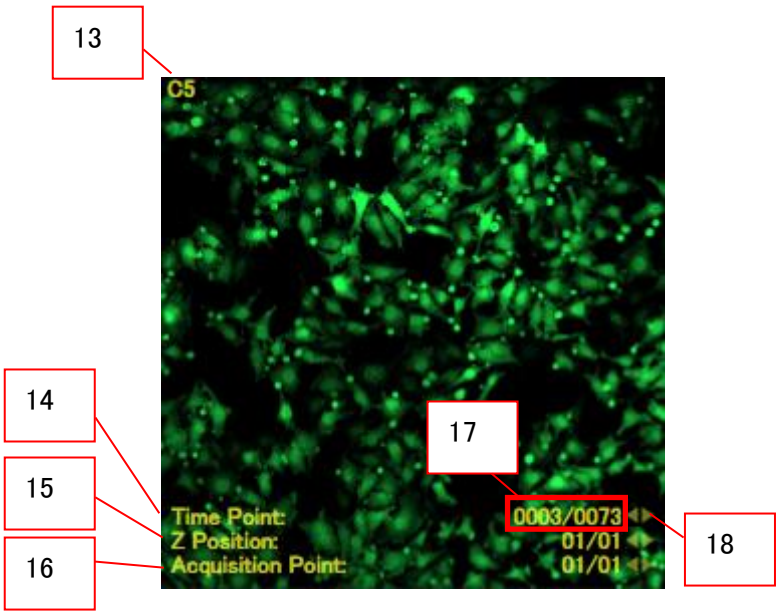
3) Select the well to display the images of.
4) Select all wells.
5) Delete all wells.
6) Select the display format of the display area.

Item	Explanation
Row and Column Images	Show images in the well plate view.
Time Point Images	Show images along the time axis.
Z Images	Show images for each Z slice.
Acquisition Point Images	Show images for each acquisition point.

7) Show/hide channel images.
8) Adjust the contrast/change the display color.
9) Setting of the maximum intensity for contrast adjustment
10) Display area
11) Select the display format for channel images.

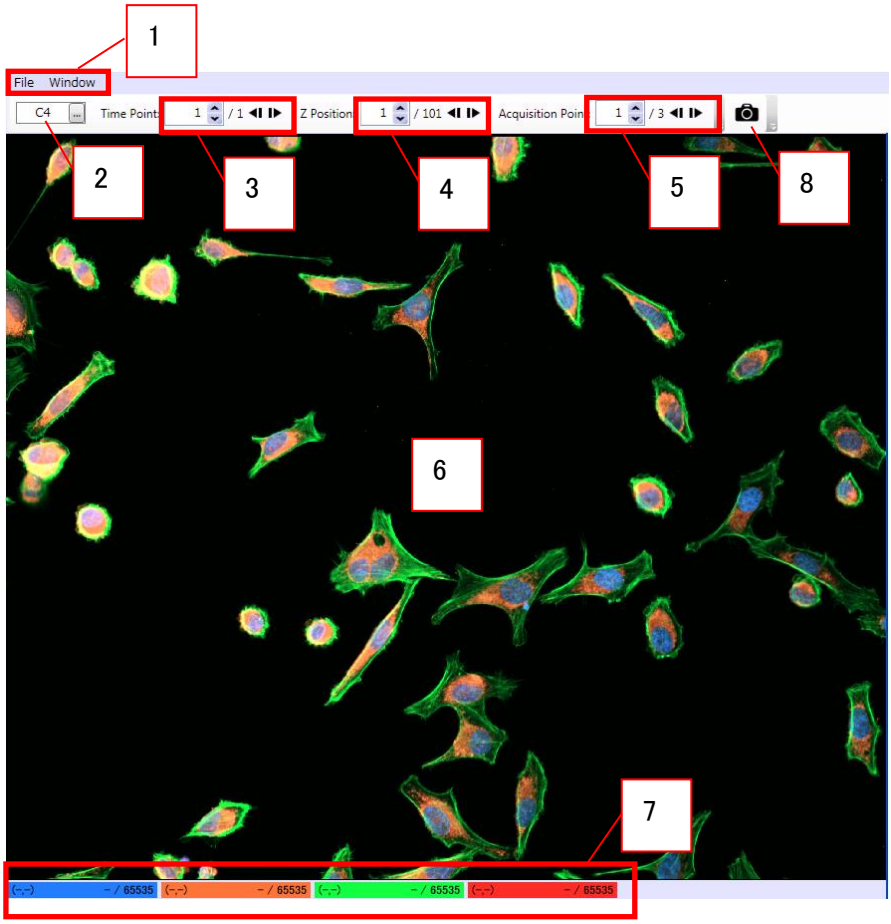


12) Change the number of all images displayed on the display area.
(Image number of the image being displayed/total number of images)



13) Well number
14) Time series
15) Z position
16) View field
17) Image number of the image being displayed / total number of images
18) Show the next image.

Measurement Result Sub Screen



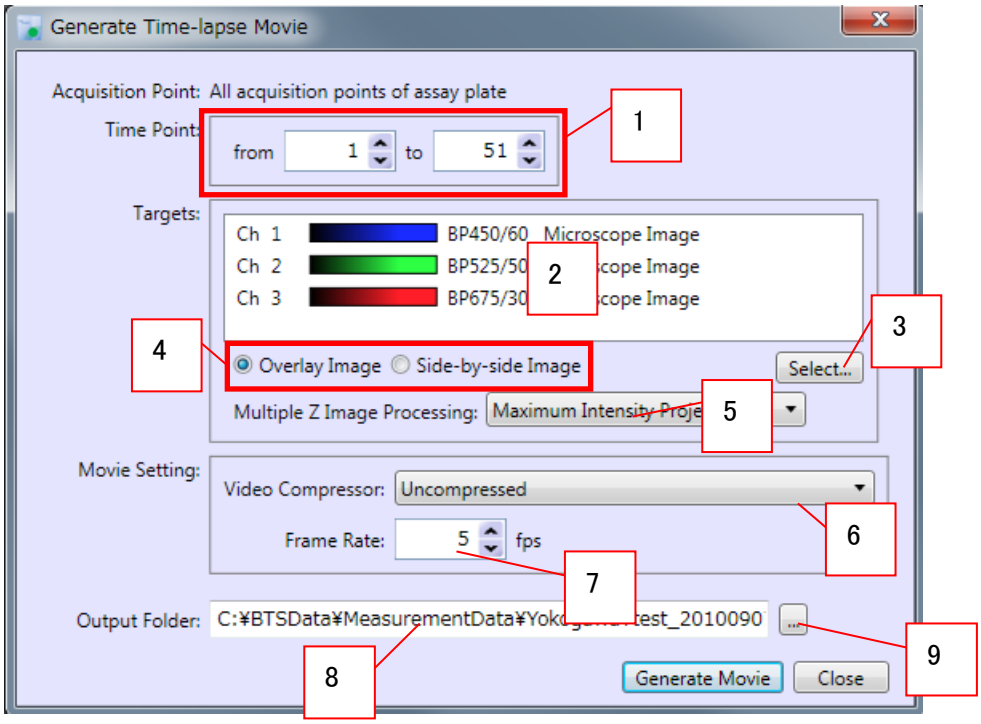
1) Menu

File menu	Explanation
Generate Movie	Display the screen to create movie files.
Save Snapshot	Capture a snapshot of image being displayed.
Close	Close the measurement result display screen.

Window menu	Explanation
Maximize	Display measurement result sub screens by maximizing.
Restore	Restore measurement result sub screens.
Cascade	Display measurement result sub screens by cascade mode.
Tile	Display measurement result sub screens by tiling.
Close All Image Windows	Close all measurement result sub screens.

2) Change the well to display.
3) Change the time point to display.
4) Change the Z position to display.
5) Change the field to display.
6) Display area
7) The pixel position and intensity of each channel
8) Capture a snapshot of image being displayed.

Generate Time-lapse Movie Screen



1) Specify the time point range to create movie files.
2) The channels to create movie files
3) Select the channels to create movie files.
4) Output items for channel images

Item	Explanation
Overlay Image	When creating movie files, channel images are output by overlaying.
Side-by-side Image	When creating movie files, channel images are output by tiling.

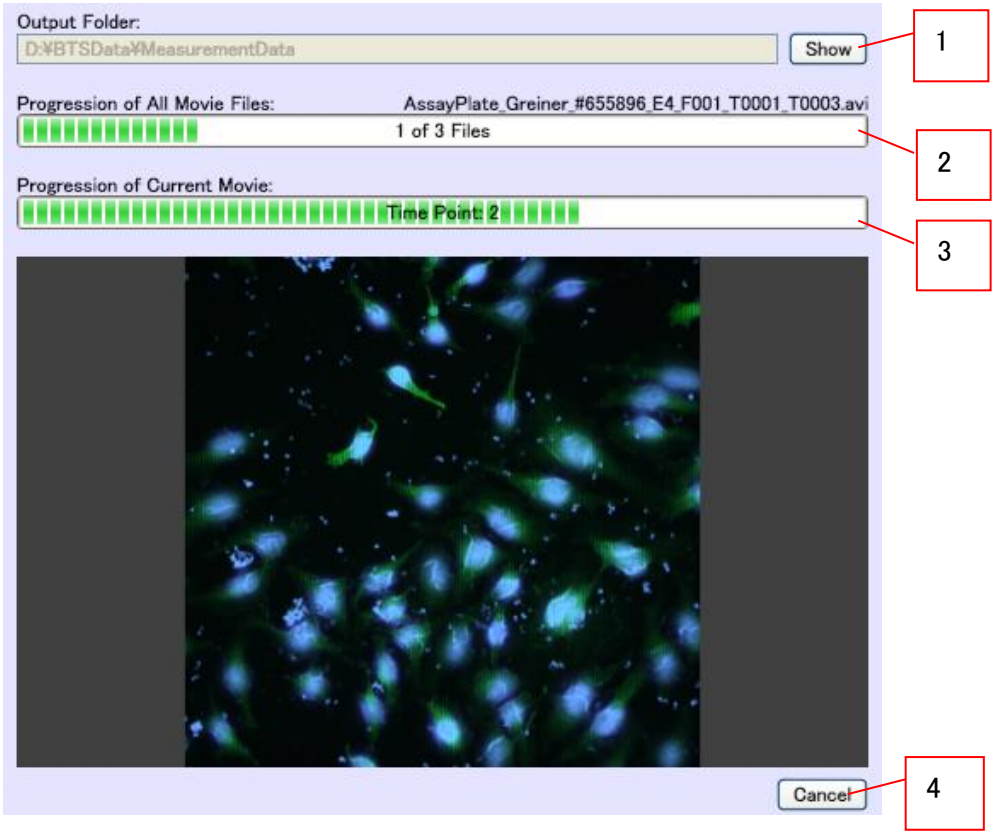
5) Select an output method for Z-stack images.

Item	Explanation
None	No Z-stack images exist.
Maximum Intensity Projection	Create movie files by MIP images. (Refer to 5.1.)
Minimum Intensity Projection	Create movie files by MinIP images. (Refer to 5.1.)
Average Intensity Projection	Create movie files by AIP images. (Refer to 5.1.)
Maximum Intensity Image	Create movie files using the Z images which have the highest total intensity in the Z-stack images at each time point.
Sum Intensity Pro-jection	Create movie files by SUM images. (Refer to 5.1.)

6) Select a compression method for creating movie files.
7) Specify the number of images replaying per second.

8) Folder path to output movie files
9) Select a folder to output movie files.

Generating Time-lapse Movie Screen

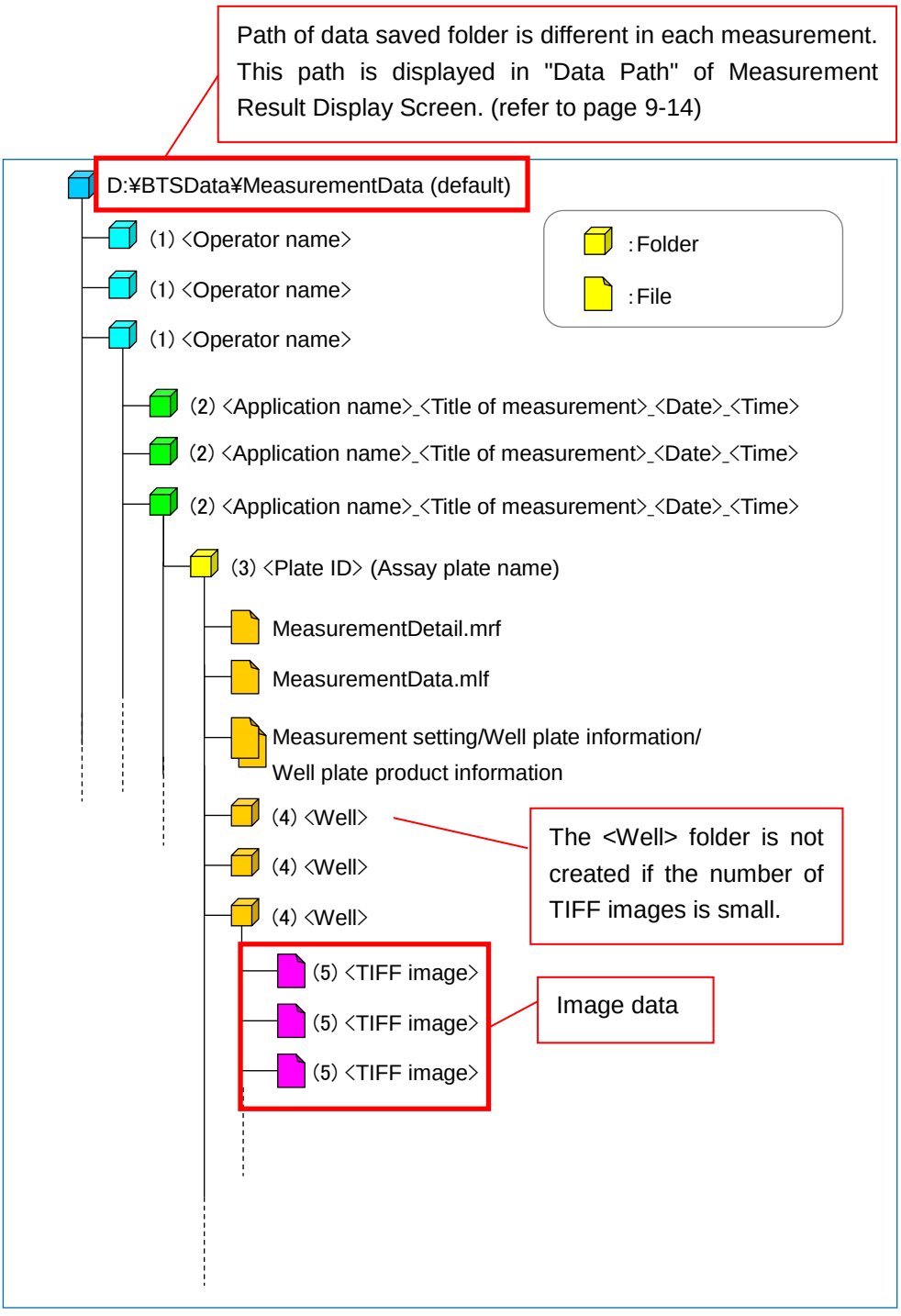


1) Display the output folder.
2) Processing bar for creating all movie files
3) Processing bar for creating current movie file
4) Stop creating movie files.

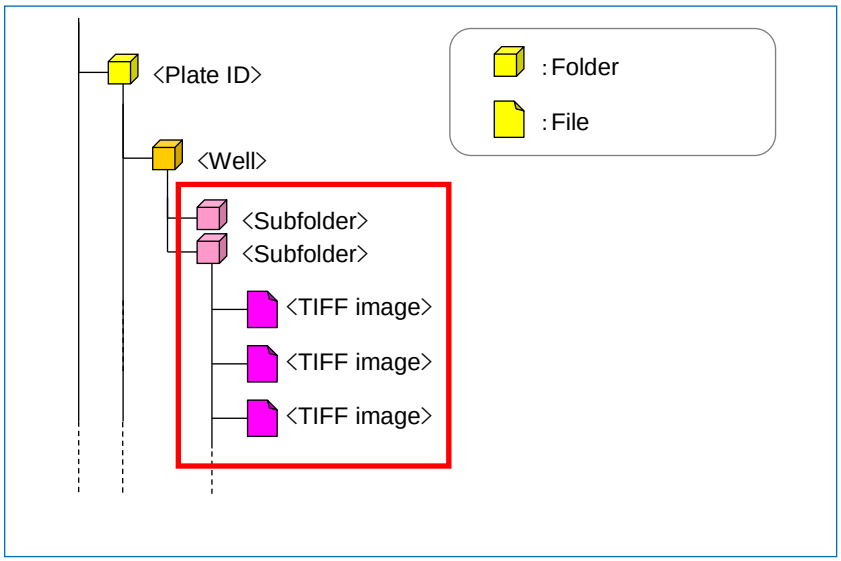
10. Saving Measurement Files

10.1. Save Location of Measurement Files

Folder for measured data has the following hierarchical structure.



If there is a large amount of data, multiple subfolders are created and image files are divided and saved in these sub-folders.



10.2. Image File Name

Image files are saved in the following format:

<Plate ID>_<Well>_T<tpIndex>F<fpIndex>L<tlIndex>A<alIndex>Z<zpIndex>C<chIndex>.tif

Plate ID : Assay plate name
Well : A1, H12, etc.
tpIndex : Time point number (0001 to 9999)
fpIndex : Imaging point number (001 to 999)
tlIndex : Time line number (01 to 99)
alIndex : Action List number (01 to 99)
zpIndex : 3D Z number (01 to 99)
chIndex : Imaging channel (01 to 99)

11. Troubleshooting

11.1. Trouble Items

Condition	Check item
The power does not turn on.	Is the power plug connected to the power outlet?
	⇒Check the connection.
	Is 230 VAC power supplied to the power outlet to which the main unit is connected?
	⇒Check the breaker installed in the room or other applicable location in the building.
	Is the main power switch on the main body turned on?
	⇒Turn on the main power switch at the back of the main unit.
The dispenser does not operate or reagent does not drip.	Does the source plate contain reagent?
	⇒Put reagent in the source plate.
	Isn't the source plate set in a wrong orientation on the shuttle?
	⇒Set the plate so that the well A1 comes to the top left-hand corner of the shuttle.
	Have you entered a well plate information file?
	⇒Enter an appropriate value under "Reagent Volume" in the well plate information file.
	Are the correct well plate products selected?
	⇒When entering information in the well plate information file, well plate products are selected. Select appropriate well plate products. If a wrong product is selected, the dispenser may be damaged.
	Isn't the tip rack set in a wrong orientation?
	⇒The tip rack set on the tip platform should be oriented so that remaining tips face the left side toward you.
	Have you entered a dispensing setting file?
	⇒Enter appropriate values under "Aspirate" and "Dispense." It is recommended that you perform simulation once.
	Have you set "Dispense Operation" in the action list area of the measurement setting file and specified a dispensing setting file?
	⇒Specify the correct dispensing setting file under "Dispense Operation."

Condition	Check item
The dispenser does not operate or reagent does not drip.	Do the acquisition points under “Dispense Operation” match the dispensing points in the dispensing setting file?
	⇒Dispensing is not performed unless there are wells matching the acquisition points specified under “Dispense Operation” in the measurement setting file and dispensing points specified in the dispensing setting file.
Images cannot be measured.	Isn't the bottom of the well plate dirty?
	⇒If the bottom of the well plate is dirty with attachment of dust or oil, a focus error may occur. Use a cloth, etc., to thoroughly remove the soiling.
	Are the selected well plate products correct?
	⇒When entering information in the well plate information file, well plate products are selected. Select appropriate well plate products.
	Isn't the assay plate set in a wrong orientation on the stage?
	⇒Set the assay plate so that the well A1 comes to the top left-hand corner of the stage.
Automatic analysis can not be performed.	Is the CV7000 Analysis Software launched?
	⇒To display the AnalysisProtocol setting in the Start Measurement screen, launch the CV7000 Analysis Software and set to communicate the net-work previously.
Images are out of focus.	Have you set software focus?
	⇒When software focus is set, the cell plane to be captured becomes stable and the focus accuracy improves.
Images are dark or otherwise cannot be seen.	Isn't the exposure time too short?
	⇒Set a longer exposure time.
	Isn't the laser too weak?
	⇒Raise the laser output.
	Isn't the staining time too short?
	⇒Dark samples can be captured brighter by setting the binning more than "2x2."
	Isn't the filter set or laser selected incorrectly?
	⇒Set the imaging channels correctly.
The focus shifts in time-lapse imaging.	Is temperature control implemented?
	⇒Control the plate heater temperatures because a temperature shift causes the focal plane to deviate. It takes about 30 minutes after the plates are set on the stage until the temperatures stabilize.

Condition	Check item

Patterns exist in image	Are there water drops on bottom of wellplate?
	⇒Water drops are attached on bottom of wellplate by temperature difference and they can be imaged. Wipe the water drops by soft cloth and others.
AF (auto focus) error occurred.	Isn't the well plate bottom too dirty?
	⇒Wipe the plate bottom so as to be clean. AF error will occur if droplet or dirt is attached onto well plate bottom.
	Is the selected well plate product correct?
	⇒When entering information in the well plate information file, well plate products are selected. Select appropriate well plate products.
	Isn't the well plate inclined?
	⇒Remove well plate from the stage, and reset the plate on the stage.
	Don't you observe around well edge?
	⇒Sometimes, It is difficult to auto-focus around well edge because of adhesive material. It is recommended to observe around well center.
	Is well filled with liquid solution?
	⇒In the case of drying in well, it may cause auto-focus error. Fill well with liquid solution.
	Don't you observe circumference wells when preview?
	⇒Depending on the selection of objective lens and well plate product, it may cause AF error around well area of plate circumference. It is recommended to use long working distance objective lens such as “20x Long W.D.”.
	Don't you select wrong lens type for water immersion lens or dry lens?
	⇒Select each available lens type correctly for water immersion lens or dry lens. (Refer to 5.14.)

11.2. Message List

Warning Message	Description
Camera <camera> is not stably cooled: <temperature> ℃	Camera is not enough cooled. Please wait a few minutes.
Disk drive free space becomes less than present 50% after measurement.	Please delete unnecessary measurement data in D:¥BTS-Data¥ Measurement folder.
Image acquisition timed out: <cameras>	The cameras do not respond until timeout. The measurement software automatically retries the image acquisition.
Reader device is being controlled.	Please wait for completion of the device control.
Reader device is locked for measurement.	The measurement is running. Please wait for completion of the measurement.

Unexpected System Error and Internal Program Error	Description
The following error messages are left out of this list. - Unexpected System error - Internal program error Please try actions on Description. If you can not solve the problem, please report to the support engineer.	1. Check the cables between the PC and the reader device. 2. Check the power state of the reader device. 3. Check the D:¥BTSData folder. Open this folder with Windows Explorer and confirm that the files can be accessed. 4. If the error message includes the detail code such as '??_MANU,<id>,<error-code>,<error-message>', please check the error codes of the reader firmware. 5. Check the installation of runtime libraries. (Camera Control Software, DirectX, Visual C++ 2008 SP1) 6. Check the configuration of the PC. (IP address, firewall, local security policy, and so on)

Error Message	Description
"Lock pages in memory" privilege not assigned.	Please read the installation guide and change local policy through Windows control panel.
<laser wavelength> lasers are not turned on.	Please turn lasers on using 'Device Console' -> 'Laser'.
<number> camera devices not detected	Please check the cables between the PC and the reader device, and check the power state of the reader device.
<wellplate information> is not <number> wells.	The number of wells of the selected wellplate information is not same as the number of wells of the measurement setting. Please select the same number of wells of wellplate information as the measurement setting.
Acquisition Point Error at <well>: (position)	The reader device can not move to the specified position of the well. The measurement continues when this error occurs.
AF Error at <well> No. <number> (<assay plate name>)	The auto focus unit can not detect the bottom of the wellplate. Please check that the correct wellplate is loaded. If the bottom of the wellplate is not clear, the auto focus unit may cause an error. The measurement continues when this error occurs.
AF Error occurred.	The auto focus unit can not detect the bottom of the wellplate. Please check that the correct wellplate is loaded. If the bottom of the wellplate is not clear, the auto focus unit may cause an error.
AF parameter not found: <wellplate name> (<objective>)	You must obtain the auto focus parameter for the specified wellplate and the specified objective.
Corrected image data directory not found	Please check D:¥BTSData¥CorrectedMeasurementData folder.
Directory access failed, <folder>	Please check the specified folder. It may not exists.
Dispense operation is not available on multiple time points.	Multiple dispensing is not supported. Please delete other 'Dispense Operation' action.
Dispense Timing must be less than Period.	The value of dispense timing exceeds the period of the live imaging. Please enter the valid dispense timing.
Elapsed time exceeds the next time point.	The elapsed time of the measurement exceeds the expected measurement time. Please open the measurement setting and save it again.
Exposure Time of Ch <number> is greater than <number> ms.	This error occurs when the exposure time is less than <number> ms. (This error message is inappropriate. It will be changed.) Plaese increase the value of Exposure Time of the specified channel greater than <number> ms.

Exposure Time of Ch <number> is greater than Interval.	Please increase the value of Interval of Live Imaging or decrease the value of Exposure Time of the specified channel.
Failed to allocate camera buffer.	This error occurs when the free memory of the process is not enough. Please restart the measurement software.
Failed to allocate the native CameraBuffer object.	This error occurs when the free memory of the process is not enough. Please restart the measurement software.
Failed to aspirate at <well> (source plate name): <detail>	Please check the detail of the error code. If it includes '??_MANU', the reader device causes an error.
Failed to count tips.,Please set a new tip rack.	Please load the filled tip rack.
Failed to create directory: <detail>	Please check D:¥BTSData folder. This error occurs when D: drive does not exist.
Failed to dispense at <well> (assay plate name): <detail>	Please check the detail of the error code. If it includes '??_MANU', the reader device causes an error.
Failed to execute the measurement.,<detail>	Please check the detail of the error code. If it includes '??_MANU', the reader device causes an error.
Failed to get reader device status	Please check the reader device.
Failed to initialize frame grabber.	This error occurs when the free memory of the process is not enough. Please restart the measurement software.
Failed to initialize measurement software.	Please check the detail of the error code. If it includes '??_MANU', the reader device causes an error.
Failed to initialize video compressor: <detail>	The selected video compressor is not supported by the movie generator. Please select another video compressor.
Failed to load <file>	The specified file is incorrect. Please report to the support engineer.
Failed to load the measurement data: <file>	The specified file is incorrect. Please report to the support engineer.
Failed to load the measurement setting: <file>	The specified file is incorrect. Please report to the support engineer.
Failed to load the well plate product information: ID=<number>	The <number> .wpp file is incorrect. Please report to the support engineer.
Failed to lock reader device control.	The measurement software is controlling the reader device (such as start-up). Please wait for the completion of the device control.
Failed to prepare measurement. Please try again later.	The measurement software is controlling the reader device (such as start-up). Please wait for the completion of the device control.
Failed to prepare the measurement.,<detail>	The reader device is working. Please wait for the completion of the device control.
Failed to read barcode.,<detail>	Please check the barcode of the wellplate. It may not be clear.

Failed to save image. Image Processing: <image processing>	Please check the disk free space of D: drive.
Failed to save the measurement setting: <file>	Please check the disk free space of D: drive.
Failed to scan <folder>	Please check the specified folder. It may not exist.
Failed to shut down device,<detail>	Please check the detail of the error code. If it includes '??_MANU', the reader device causes an error.
Failed to start device,<detail>	Please check the detail of the error code. If it includes '??_MANU', the reader device causes an error.
Failed to test 2D acquisition.	Please check the detail of the error code. If it includes '??_MANU', the reader device causes an error.
Failed to test 3D acquisition.,<detail>	Please check the detail of the error code. If it includes '??_MANU', the reader device causes an error.
Failed to test acquisition parameters.,Measurement is running.	You can not use the image acquisition test when the measurement is running. Please wait for the completion of the measurement.
Failed to update data directory: <detail>	Please check D:¥BTSData folder. This error occurs when D: drive does not exist.
Image acquisition timed out.	The camera does not respond until timeout. Please retry acquire image.
Insufficient disk drive free space.	This error occurs when D:¥BTSData¥MeasurementData folder contains many image files. Please delete unnecessary measurement data.
Insufficient memory.	This error occurs when the free memory of the process is not enough. Please restart the measurement software.
Interval of live option is less than <number> ms.	Please increase the value of Interval of Live Imaging.
Invalid application name.	You entered the incorrect application name. For example, the following characters are not available: ¥ / : * ? " < > |
Invalid density.	You entered the invalid value of the CO2 density. Please enter the value from 0.0 to 9.9.
Invalid filter of Ch <number>.	You do not select the filter of Acquisition menu of the channel setting. Please select an item of Acquisition menu.
Invalid flow rate.	You entered the invalid value of the CO2 flow rate. Please enter the value from 0 to 500.
Invalid measurement setting. Please check it.	The measurement setting file is incorrect. Please report to the support engineer.
Invalid number formats of operating system. Decimal symbol must be ".". Please change number formats of operating system with Control Panel. Program will be exited.	The supported decimal symbol is only ".". Please check the regional formats of Windows: 'Start Menu' -> 'Control Panel' -> 'Region and Language' -> 'Formats' If 'Format' is not 'English', please change the format.

Invalid parameter specified.	Please check the input parameter. Invalid parameter field is marked with red color.
Invalid path of Output Folder.	You entered the invalid path name. Please enter the valid full path name of the output folder.
Liquid surface detection failed at <well> (<source plate name>)	Please check the detail of the error code. If it includes '??_MANU', the reader device causes an error.
Live Imaging is not available on time lapse.	You can not specify the interval time on 'Time-lapse Setting' when 'Live Imaging' of 'Fluorescence Acquisition' or 'Bright-field/Phase-contrast' is turned on.
Loaded tip rack is not available.	Please load the filled tip rack.
Local IPv4 network address not found: Local host=<hostname>	Please check the configuration of IP address.
Measurement is already running.	Please wait for the completion of the measurement.
No image data exists.	This error occurs when you generate the movie with the channel which contains no image file. Please deselect such the channel.
No Image Processing selected.	Please select an item of Image Processing.
No objective selected.	The objective lens is not selected on the channel setting of the measurement setting. Please select the objective lens.
No reagent volume for <well> <source plate name>	The values of the reagent volume are not specified in the wellplate information. Please enter the reagent volume in the wellplate information.
No sample volume for <well> <assay plate name>	The values of the sample volume are not specified in the wellplate information. Please enter the sample volume in the wellplate information.
No target is selected.	Please select targets.
No Video Compressor selected.	Please select an item of Video Compressor.
Not enough tips to start measurement. Please set a new tip rack.	Please load the filled tip rack.
Operator, Measurement Title, or Assay Plate name is too long.	You entered long operator name, long measurement title, or long assay plate name. Please shorten the length of them.
Output folder must be full path.	You entered the relative path name. Please enter the valid full path name of the output folder.
Please add a channel.	This error occurs when you delete the channels of the measurement setting. Please add a channel.
Please add a time line and set its parameters.	This error occurs when no time line exists on the measurement setting. Please add a time line.

Please enter correct parameter.	Please check the input parameter. Invalid parameter field is marked with red color.
Please enter correct power of <laser>.	You entered the invalid value of the laser/lamp power. Please enter the value from 0 to 100.
Please enter dispense setting name.	On 'Dispense Operation' action, You do not enter the dispense setting name.
Please enter valid file name.	You entered the incorrect file name. For example, the following characters are not available: ¥ / : * ? " < > |
Please enter valid parameters.	Please check the input parameter. Invalid parameter field is marked with red color.
Please put <assay plate, source plate, or tip rack>.	The assay plate, the source plate, or the tip rack is not loaded. Please load them.
Please select a well plate type.	You do not select a wellplate type on the list view. Please select one.
Please select an item.	You do not select an item on the list view. Please select an item.
Please select the bright-field/phase-contrast target.	On 'Bright-field/Phase-contrast Acquisition' action and 'Z-Stack Bright-field/Phase-contrast Acquisition', you must select a 'Brightfield' channel or an 'Phase Contrast' channel.
Please select the different Acquisition of Ch <number>.	You selected the two or more channels which can not be acquired at a time. To acquire the channels at a time, the following parameters must be different from each other: Acquisition.
Please select the fluorescence targets.	On 'Fluorescence Acquisition' action and '3D Fluorescence Acquisition', you must select a 'Confocal Fluorescence' channel or an 'Epifluorescence' channel.
Please select the same Method, Objective, Light Source of Ch <number>	You selected the two or more channels which can not be acquired at a time. To acquire the channels at a time, the following parameters must be unique: Method, Objective, Light Source.
Please select the target.	You do not select a channel. Please select.
Reader is working now.	The measurement software is controlling the reader device (such as start-up). Please wait for the completion of the device control.
sCMOS camera not found.	Please check the cables between the PC and the reader device, and check the power state of the reader device.
The action contains invalid parameters.	Please check Action List of the measurement setting. Action shows the reason of the error.
The dispense operation already exists in the measurement setting.	You can specify only one dispense operation on the measurement setting. Please delete the other dispense operation.
The number of the channels must be up to 99.	This error occurs when you add more than 99 channels to the measurement setting. Please delete unnecessary channels.

The reader device is working now.	The measurement software is controlling the reader device (such as start-up). Please wait for the completion of the device control.
The selected target is not bright-field/phase-contrast.	On 'Bright-field/Phase-contrast Acquisition' action and 'Z-Stack Bright-field/Phase-contrast Acquisition', you must select a 'Brightfield' channel or a 'Phase Contrast' channel.
The selected target is not confocal.	On 'Software Focus' action, you must select a 'Confocal Fluorescence' channel.
The selected target is not fluorescent.	On 'Fluorescence Acquisition' action and '3D Fluorescence Acquisition', you must select a 'Confocal Fluorescence' channel or an 'Epifluorescence' channel.
Time Line: <time-line> Action List contains invalid parameter.	Action List of the measurement setting contains invalid parameter. Please check Action List. Invalid parameter field is marked with red color.
Time Line: <time-line> Time-lapse Setting contains invalid parameter.	Time-lapse Setting of the measurement setting contains invalid parameter. Please check Time-lapse Setting. Invalid parameter field is marked with red color.
Time Line: <time-line> Well Plate Scan Setting contains invalid parameter.	Well Plate Scan Setting of the measurement setting contains invalid parameter. Please check Well Plate Scan Setting. Invalid parameter field is marked with red color.
Too many image windows are opened.	This error occurs when you open the new image window without closing other image window. Please close unnecessary image windows.
Too many targets are selected. Please select up to 4 targets.	You selected more than 4 targets. Please select up to 4 targets.
You cannot test acquisition parameters when measurement system is working.	You cannot use the image acquisition test when the measurement is running. Please wait for the completion of the measurement.
You can select either immersion lenses or non-immersion lenses in a measurement setting.	You selected both the immersion lens and the non-immersion lens. Please select either one of them.

11.3. Error Code List

WP Error code list

Error message	Error code	Measures	Cause of the trouble
WP_IM_WIM_OBJ_CANT_USE	-204713	Select the Water immersion objective mode	Used water immersion objective lens without water.
WP_IM_DRY_OBJ_CANT_USE	-204714	Select the dry objective mode	Used the dry objective with Water immersion objective mode
WP_IM_WA_SPLY_NOT_READY	-204715	Turn on the water supply system.	Used the Water immersion objective without water supply system.

CD Error code list

Error message	Error code	Measures	Cause of the trouble
CD_PLATE_INFO_ERR	-206311	Make sure the source plate information	Source plate information is incorrect.
CD_WELL_POS_PARAM_ERR	-206313	Make sure the source plate information Set the dispensing position at center of the well.	Dispensing position is out of range
CD_ARM_XY_NOT_ESC	-206320	Return Z motor to origin position	Dispenser is not at the evacuation site.
CD_ARM_Z_NOT_ESC	-206321	Return Z motor to origin position	Dispenser is not at the evacuation site.
CD_CURR_ARM_POS_ERR	-206323	Set the dispenser at the center of the well.	Dispenser is not at the center of the chip.
CD_WELL_LIQ_VOL_ERR	-206340	Make sure the plate information Check the filling and dropping volume.	Volume setting of filing and dropping is invalid.
CD_TIP_ALREADY_PICKUP	-206350	Remove the chip from the dispenser	Performed chip count/mount while the chip already existing on the dispenser.
CD_FAIL_TO_TIP_EJECT	-206351	Re-register the chip mounting position Make sure that dump has not jammed.	CV7000 failed to discard the chip.
CD_TIP_NOT_PICKUP	-206352	Try again by mounting the chip Re-register the chip mounting position	Chip was not mounted on the dispenser.
CD_FAIL_TO_TIP_PICKUP	-206353	Re-register the chip mounting position	Failed to pick up the chips

CD_FAIL_TO_TIP_DETECT	-206354	Re-register the chip mounting position	Chip was mounted during the chip detection.
CD_TIP_CNT_UNKNOWN	-206355	Count the chip number.	CV7000 was not sure the number of remaining chips.
CD_TIP_USABLE_CNT_IS_0	-206356	Replace the chip rack	Number of chips at the chip rack was to empty
CD_TIP_RACK_UNUSABLE	-206359	Make sure that the incorporation of the chip rack	Chip rack is not mounted
CD_TIP_CHUTE_DUCT_JAM	-206360	Make sure that dump has not jammed.	Chip may jammed in the dump

MS_IW Error code list

Error message	Error code	Measures	Cause of the trouble
MS_IW_POWER_OFF	-201211	Turn ON the power of the CV7000	CV7000 power is not turned on
MS_IW_EMERGENCY	-201214	Turn OFF the emergency stop button.	Emergency stop button is turned ON
MS_IW_AIR_SUPPLY_ERR	-201223	Supply the air to CV7000	Air is not supplied to the CV7000

LS Error code List

Error message	Error code	Measures	Cause of the trouble
LS_INTERLOCK_ERR	-207016	Cancel the interlock by using adjustment tools	Interlock shutted down the laser for safety.
LS_ERR_LS_NOT_READY	-207041	Launch the laser.	Manipurated the laser power while laser has not ready.

ST Error code list

Error message	Error code	Measures	Cause of the trouble
ST_PLATE_INFO_ERR	-203010	Make sure the plate information	Plate information is incorrect.
ST_TRGT_WELL_NUM_ERR	-203012	Set the stage movement within well radius	Stage movement has variance with the well plate information.
ST_FORBIDDEN_TRGT_WELL	-203014	Do not move to specified well	Stage has moved to Unavailable area
ST_FORBIDDEN_TRGT_POS	-203015	Do not move to the the specified location	Stage has moved to Unavailable area
ST_SET_SAFEGUARD	-203022	Remove the obstacles on the stage	Safety system.
ST_LIFTER_ST_POS_ERR	-203025	Re-register "Loader Handover"position. Change the position of the Sensor enable to operate the lifter	Lifter sensor could not detect the lifter.

ST_LIFTER_PLATE_EXIST	-203026	Remove the plate on the lifter.	Plate is placed on the lifter
ST_FORBIDDEN_TRGT_AREA	-203028	Do not move at the specified location	Stage has moved to Unavailable area

HT Error code List

Error message	Error code	Measures	Cause of the trouble
HT_OVERSCALE_ERR	-203521	Check wiring connections Clean the connector	The sensor detects the temperature is too high
HT_BURNOUT_ERR	-203525	Check wiring connections Clean the connector	Poor Connection Wiring is snapped

YP Error code list

Error message	Error code	Measures	Cause of the trouble
YP_MT_NOT_READY	-101001	Motor return to the origin	Motor does not return to the origin. but If you want to return to origin MTWP01 or MTWP02, please check the valve
YP_ACK_ERR	-101004	Cancel the alarm and correct the cause of the alarm	Alarm has occurred
YP_END_ERR	-101005	Cancel the alarm and correct the cause of the alarm	Alarm has occurred

12. Maintenance and Inspection

- This equipment uses many plastic parts. When cleaning these parts, wipe them with a dry, soft cloth. Do not use benzene, thinner or other chemical substance or detergent to clean plastic parts, as it may cause discoloration, deformation or damage.

- If the equipment malfunctions, contact us without attempting to access the inside or take any other actions to resolve the problem yourself.

- If you perform measurement with the dispensing operation, check that the tip rack disposal box is not full. The tip waste should be disposed of by the waste disposal contractor.

13. Warranty

1) The warranty period is one year from the date of installation. Failures that occur within the warranty period will be repaired for free.
2) The warranty applies only to this equipment.
3) If the following applies, repair will be charged even within the warranty period.

- Failures or damage caused by inappropriate handling or use.
- Failures or damage caused by handling, use, or storage that exceed the tolerance of the design and specifications.
- Failures or damage caused by the repairs and modifications made by the user.
- Failures or damage caused by the transportation, movement, dropping, or the like after purchase.
- Failures or damage caused by a fire, natural disaster (earthquake, storm, or flood disaster), salt damage, gas damage, and abnormal voltage.

4) Any other damage not attributable to Yokogawa is outside the scope of the warranty.
5) If you need any repairs, please consult with us.

Contact Us

Please contact the dealer inquiries about this product.

Manufacturer
International Sales Team
Life Science Center, Measurement Business HQ
Yokogawa Electric Corporation
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E-mail CSU_livecell_imaging@cs.jp.yokogawa.com

Home page http://www.yokogawa.co.jp/scanner

14. General Specification

Model	CV7000
Supported sample vessels	Wellplate (6, 24, 96, 384, 1536 wells), Slideglass
Observation mode	Confocal mode (simultaneous imaging of max.3 colors) Epi-fluorescence mode (simultaneous imaging of max.3 colors) 【Option】Bright field mode Phase-contrast mode (The latter is available for wellplates of 6/24 wells)
Output data format	Image data：16 bit TIFF, PNG; Numerical data： CSV, Original format
Excitation wavelength	405/488/532/640nm, all solid laser【Option】365nmLED
White light illumination	【Option】100W halogen lamp
Autofocus	Laser-based mode, Image-based mode
Objective lens	Max. 6 lenses are available： Automatically switchable Dry 4X, 10X, 20X, 40X Water immersion 60X Long working distance 20X Phase-contrast 10X, 20X (Only for 6/24 wellplates)
Confocal unit	Microlens-enhanced wide-view dual nipkow disk confocal scanner
Camera	Max. 3 cameras (sCMOS, Number of pixels：2560 X 2160, pixel size：6.5μm)
Stage incubator	【Option】Capable of live cell imaging. Temperature range：35 up to 40℃, CO_2 supply box (CO_2：5%, humidifier)
Dispenser	【Option】Disposable tip type
Bar-code reader	【Option】 Target codes：1 up to 2 dimension
Workstation	Dual-monitor workstation for system control Dual-monitor workstation for data analysis
Operating environment	15 up to 30℃ 10 up to 70%RH (No condensation)
Power supply	Measurement unit：Single-phase 200VAC, 2KVAmax Workstation for system control：100-240VAC, 1.5KVAmax Workstation for data analysis：100-240VAC, 1.5KVAmax
Dimension	Measurement unit：1496W X 1160D X 1650H (mm) Workstation for system control：1000W X 700D X 1200H (mm)
Weight	Measurement unit：650kg, Workstation for system control：50kg
Noise level	Less than 70 dB

14.1. MS Code

Main Unit

Model Code	Suffix Code	Option Code	Description
CV7000			High-throughput Cytological Discovery System
Camera	-C1		sCMOS one unit
	-C2		sCMOS two units
	-C3		sCMOS three units
Laser	F02		405, 488, 561, 640 nm
	F03		488 nm
	F04		405, 488 nm
	F05		405, 488, 561 nm
	F06		488, 561, 640 nm
	F07		488, 561 nm
UV	U		With UV light source
	N		Without UV light source
Bright Field / Phase contrast	B		With Bright field/ Phase contrast illumination
	N		Without Bright field/ Phase contrast illumination
Dispenser	-D		96 tip type dispenser, loader (reagent plate and tip rack)
	-E		384 tip type dispenser, loader (reagent plate and tip rack)
	-N		Without dispenser
Stage Heater / Water Immersion	F		Full option (Temperature control, Humidifier and Water immersion)
	W		Temperature control and Water immersion
	H		Temperature control and Humidifier
	T		Temperature control
	L		Water immersion
	N		Without Stage Heater and Water immersion
CO_2 Supply	C		With CO_2 supply
	N		Without CO_2 supply
Sub code	-00		Always "-00"
Power Supply	-0		AC 200 V
	-1		AC 220 V
	-2		AC 230 V
	-3		AC 240 V
Workstation for Analysis	-14		Analysis workstation and Analysis Software (1st license)
	-13		Without analysis workstation and Analysis Software
Language	-J		Japanese
	-E		English
Option		/R1	Single barcode reader
		/R2	Double barcode reader

Objective Lens

Model Code	Suffix Code	Option Code	Description
CVLNS			Objective lens
	-7000		For CV7000
Position※1	-P☐		Objective position (without heater)
	-H☐		Objective position (with heater)
Objective Lens	-L04A		4 x dry (NA=0.16)
	-L10A		10 x dry (NA=0.40)
	-L20A		20 x dry (NA=0.75)
	-L40A		40 x dry (NA=0.95)
	-L60W		60 x water immersion (NA=1.2)
	-20AL		20 x long working distance (NA=0.45)
	-L10P		10 x phase contrast (NA=0.30)
	-L20P		20 x phase contrast (NA=0.45)
Separate Order		/S	Separate Order

※1 ☐: Position 1 - 6

CSU DM, PARA DM

Model Code	Suffix Code	Option Code	Description
CV7KDM			
	-D		Dummy code
Confocal DM (Dichroic Mirror) △: Position 1 – 2		/D△01	DM D405/488/561/640
		/D△02	DM D405/488/561
		/D△03	DM D405/488/640

Image Splitting DM

Model Code	Suffix Code	Option Code	Description
CV7KCA			
	-D		Dummy code
Image Splitting DM (Dichroic Mirror) △: Position 1 – 2		/D△01	DM488
		/D△02	DM561
		/D△03	DM640

Emission Filter

Model Code	Suffix Code	Option Code	Description
CV7KFW			
	-C☐		☐: Camera port number 1 – 3
EM (Emission Filter) △: Position 1 – 5		/△01	BP445/45 for 405nm
		/△02	BP525/50 for 488nm
		/△03	BP600/37 for 561nm
		/△04	BP679/29 for 640nm
		/△05	BP447/522 for 405nm and 488nm
		/△06	BP488/568 for 488nm and 561nm

Option

Model Code	Suffix Code	Option Code	Description
CV7KPRT			
	-D		Dummy code
Option		/WS14	Analysis workstation and Analysis Software (2nd license)