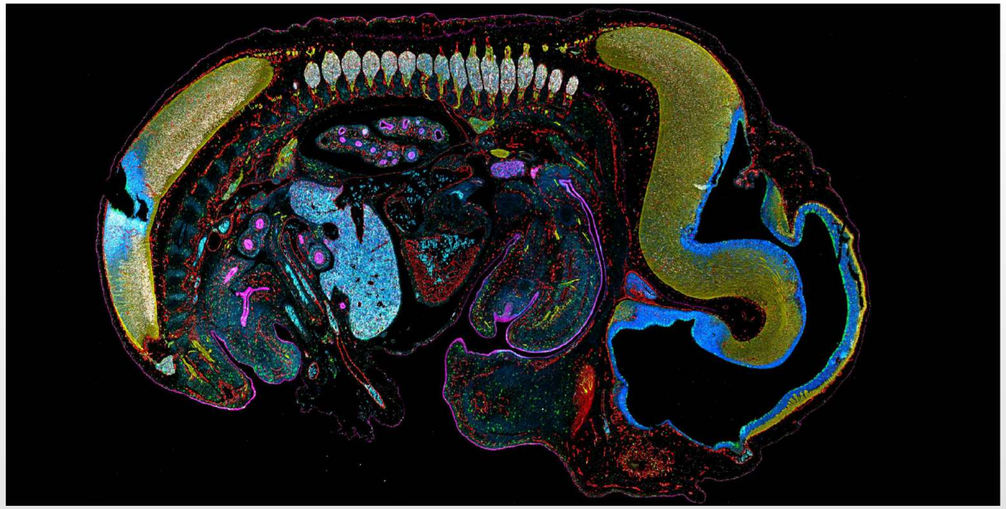
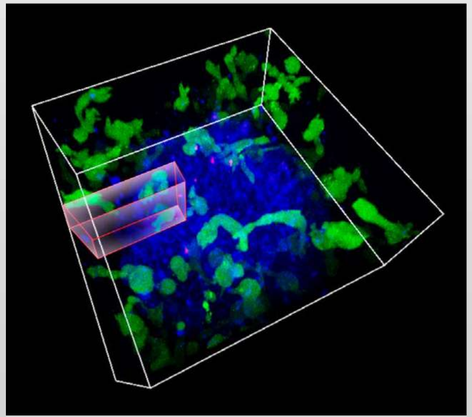
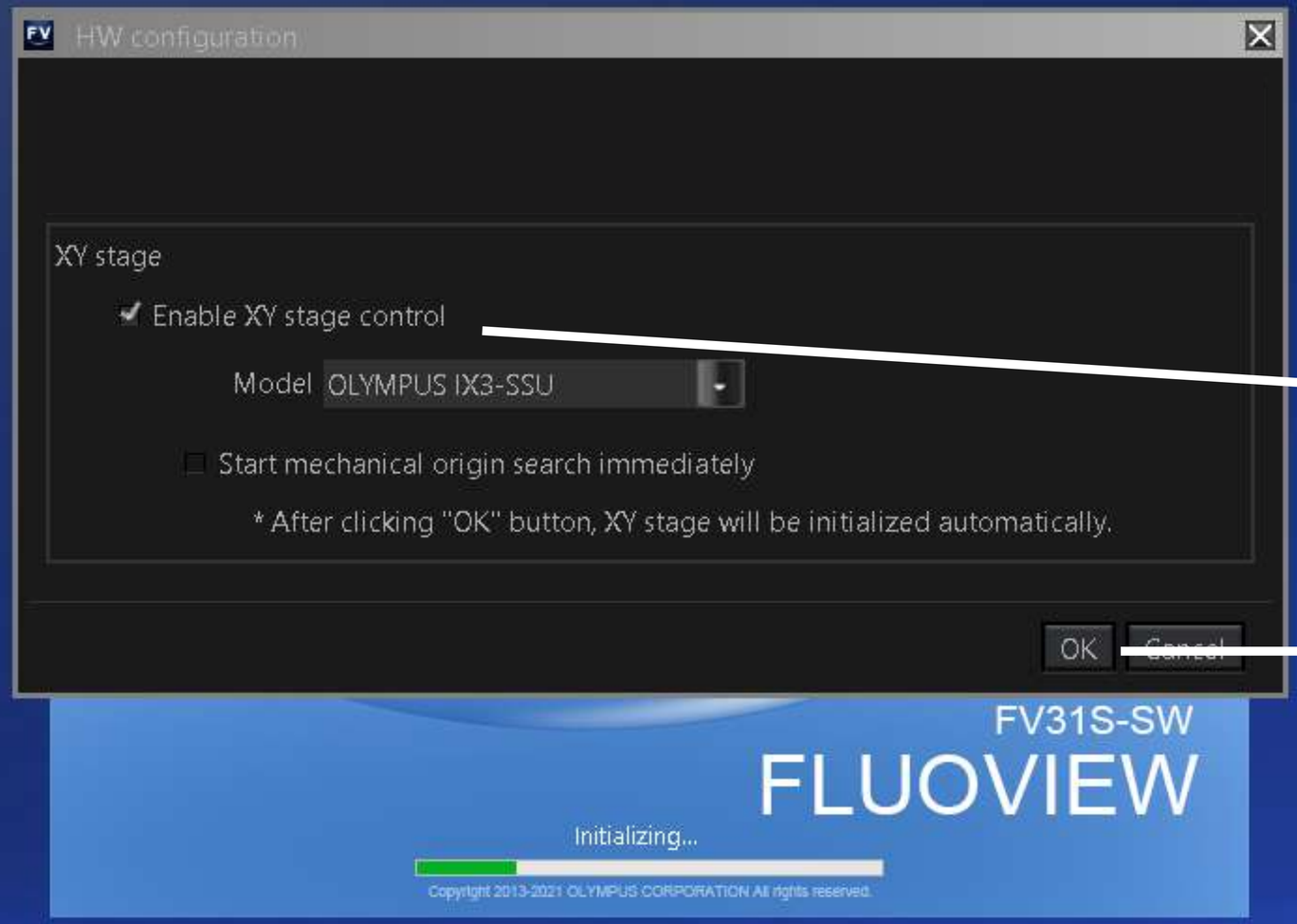
OLYMPUS FV3000 基本操作手冊



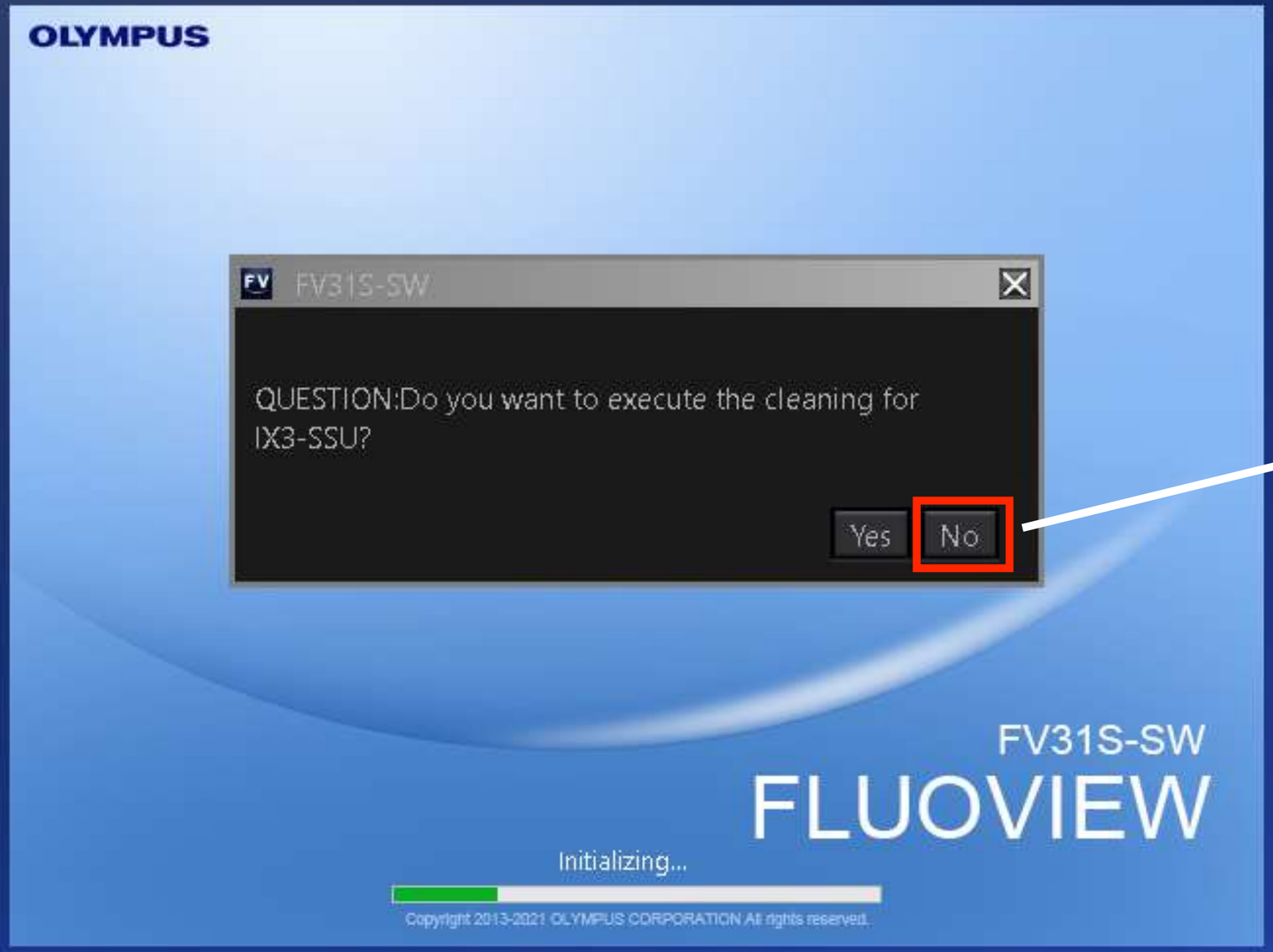



本機
資源回收筒
FV31S-DT
FV31S-SW
cellSens Dimensi...
Temp
FV3000 Manual
FV31S-SW
點選畫面上的
”FV31S-SW”
啟動軟體
OLYMPUS
英
上午 09:59
2021/7/20

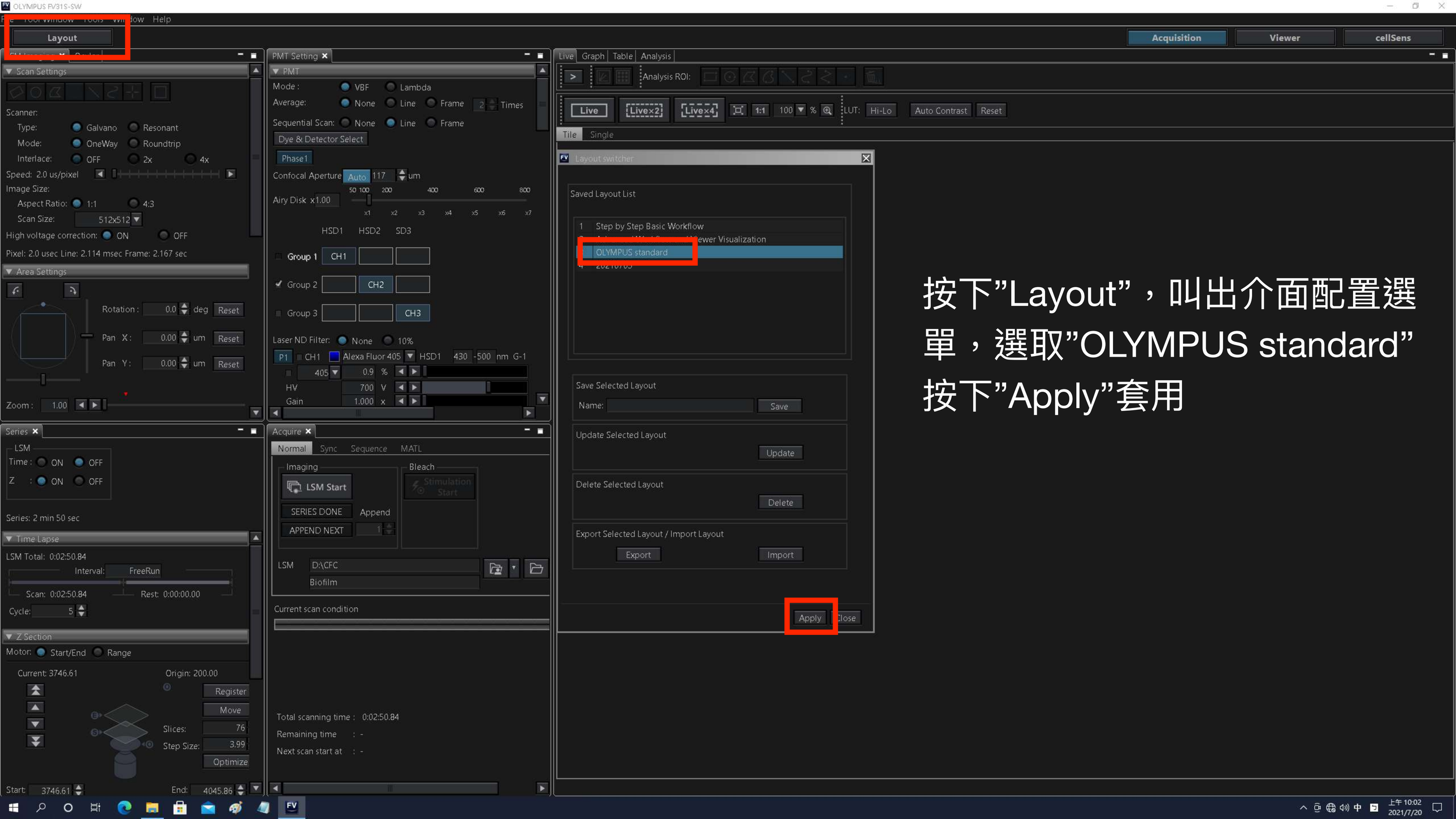


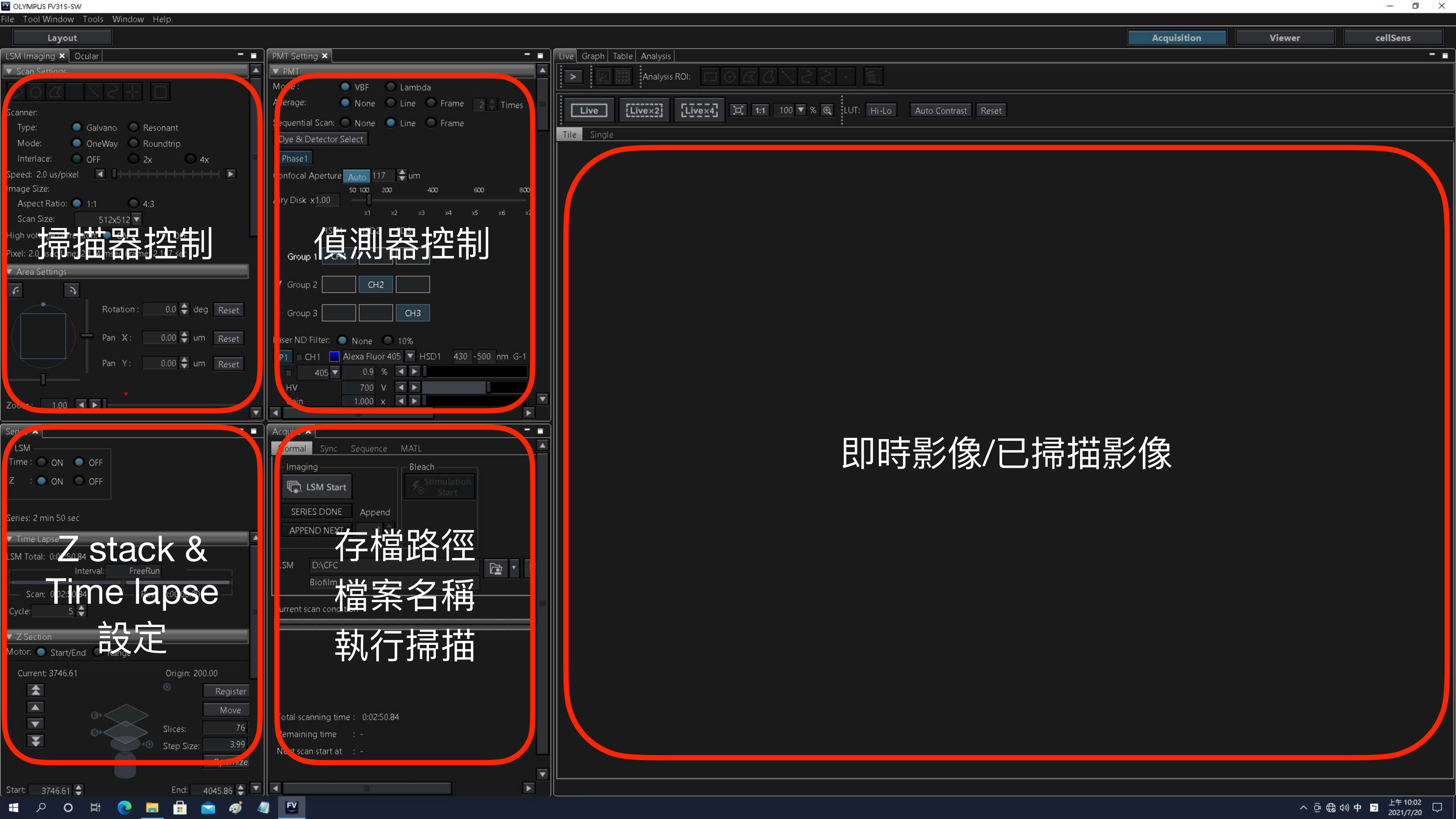
請記得勾選核取方塊
以操作電動載物臺

按下OK,進行下一步

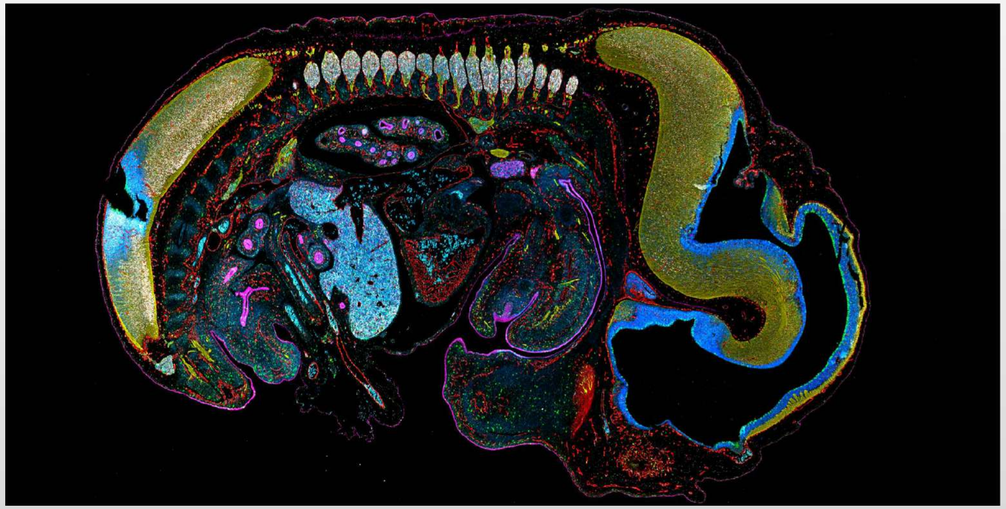


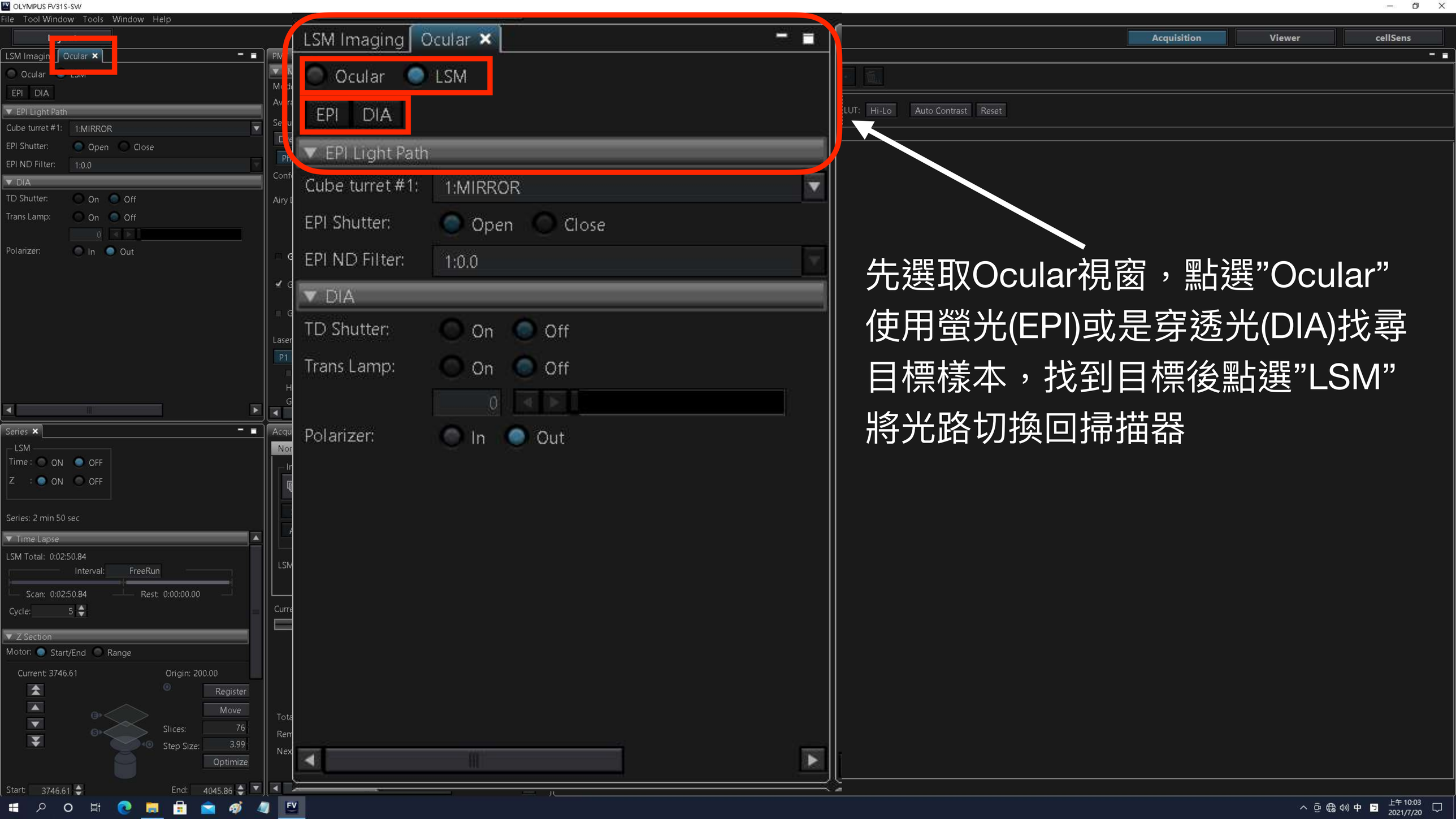
如搭配IX3-SSU電動載物臺時，請點選”No”，載物臺清潔請由工程師處理

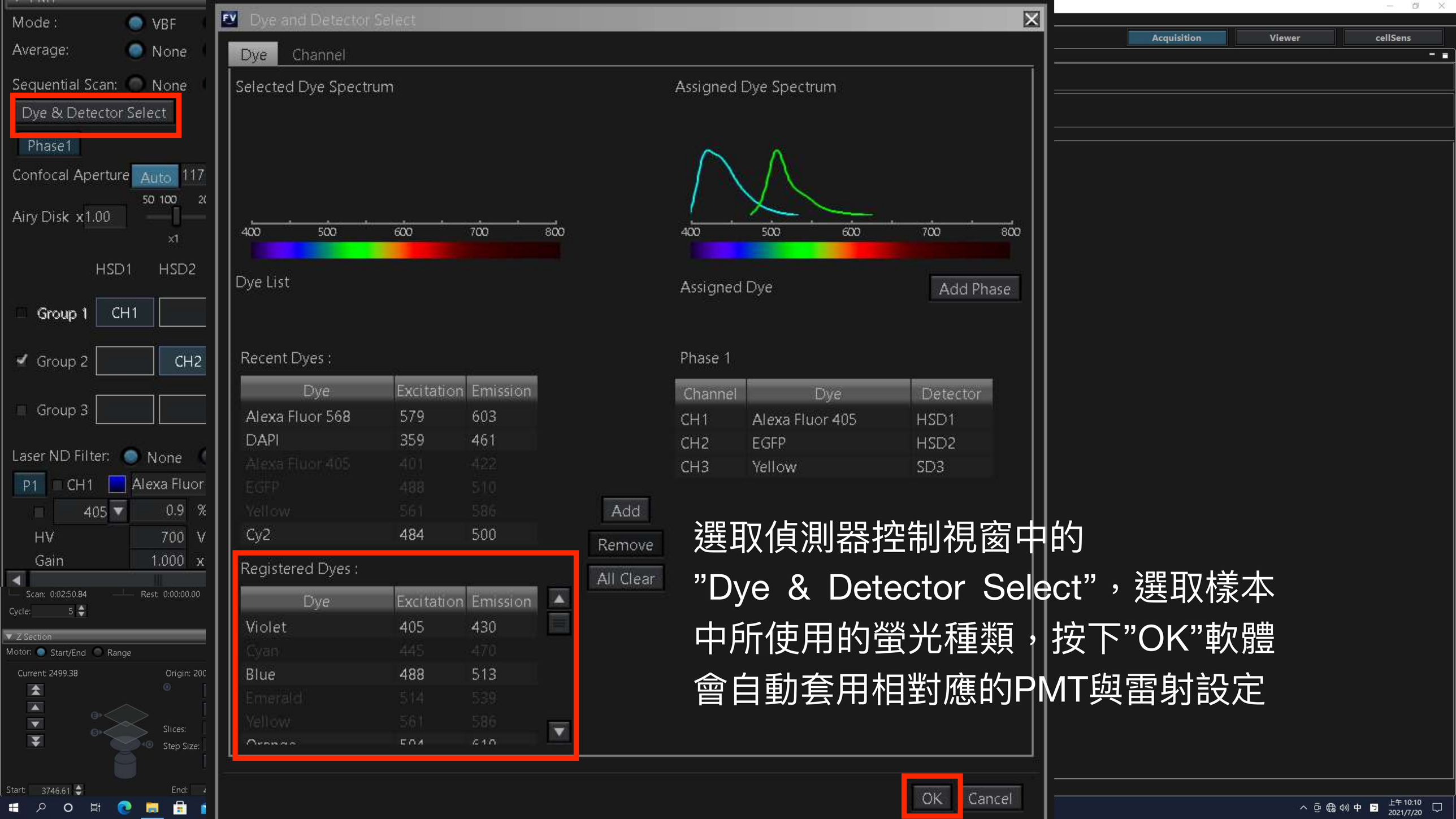

Layout
Acquisition
Viewer
cellSens
Scan Settings
PMT Setting
PMT
Live
Graph
Table
Analysis
Area Settings
Series
Time Lapse
Z Section
Acquire
Normal
Sync
Sequence
MATL
Layout switcher
Saved Layout List
1 Step by Step Basic Workflow
OLYMPUS standard
Save Selected Layout
Update Selected Layout
Delete Selected Layout
Export Selected Layout / Import Layout
Export
Import
Apply
Close
按下”Layout”，叫出介面配置選單，選取”OLYMPUS standard”
按下”Apply”套用


OLYMPUS FV31S-SW
File Tool Window Tools Window Help
Layout
Acquisition
Viewer
cellSens
LSM Imaging
Ocular
Scan Settings
Scanner:
Type: Galvano Resonant
Mode: OneWay Roundtrip
Interlace: OFF 2x 4x
Speed: 2.0 us/pixel
Image Size:
Aspect Ratio: 1:1 4:3
Scan Size: 512x512
Area Settings
Rotation : 0.0 deg Reset
Pan X : 0.00 um Reset
Pan Y : 0.00 um Reset
掃描器控制
PMT Setting
PMT
VBF Lambda
None Line Frame Times
None Line Frame
Dye & Detector Select
Phase1
Auto 117 um
Group 1 CH1
Group 2 CH2
Group 3 CH3
None 10%
CH1 Alexa Fluor 405 HSD1 430 -500 nm G-1
HV 700 V
偵測器控制
Live Graph Table Analysis
Analysis ROI:
Live Live×2 Live×4 1:1 100 % LUT: Hi-Lo Auto Contrast Reset
Tile Single
即時影像/已掃描影像
LSM
Time : ON OFF
Z : ON OFF
Series: 2 min 50 sec
Time Lapse
Interval: FreeRun
Cycle: 5
Z Section
Motor: Start/End
Current: 3746.61
Origin: 200.00
Register
Move
Slices: 76
Step Size: 3.99
Start: 3746.61
End: 4045.86
Z stack &
Time lapse
設定
Formal Sync Sequence MATL
Imaging
LSM Start
Bleach
Stimulation Start
SERIES DONE
Append
APPEND NEXT
D:\CFC
Biofilm
Total scanning time : 0:02:50.84
Remaining time : -
存檔路徑
檔案名稱
執行掃描
上午 10:02
2021/7/20

XY單張取像 Step by step

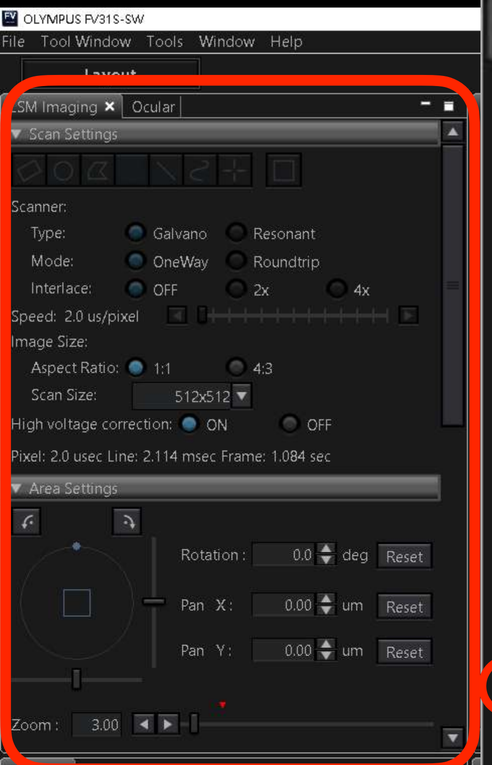


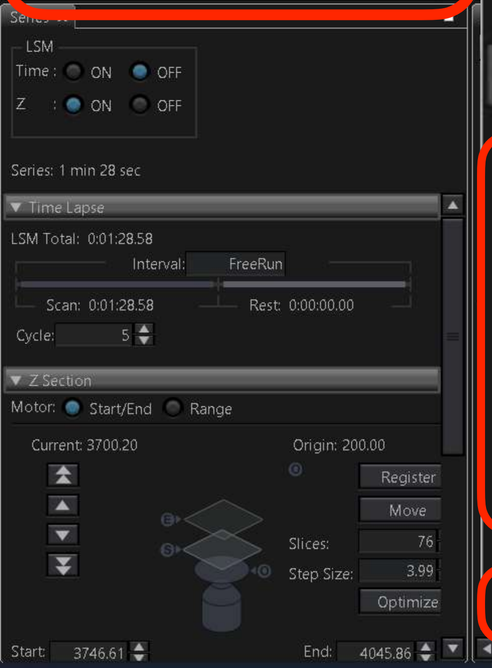

LSM Imaging
Ocular
Ocular
LSM
EPI
DIA
EPI Light Path
Cube turret #1:
1:MIRROR
EPI Shutter:
Open
Close
EPI ND Filter:
1:0.0
DIA
TD Shutter:
On
Off
Trans Lamp:
On
Off
0
Polarizer:
In
Out
Acquisition
Viewer
cellSens
Hi-Lo
Auto Contrast
Reset
先選取Ocular視窗，點選”Ocular”
使用螢光(EPI)或是穿透光(DIA)找尋
目標樣本，找到目標後點選”LSM”
將光路切換回掃描器

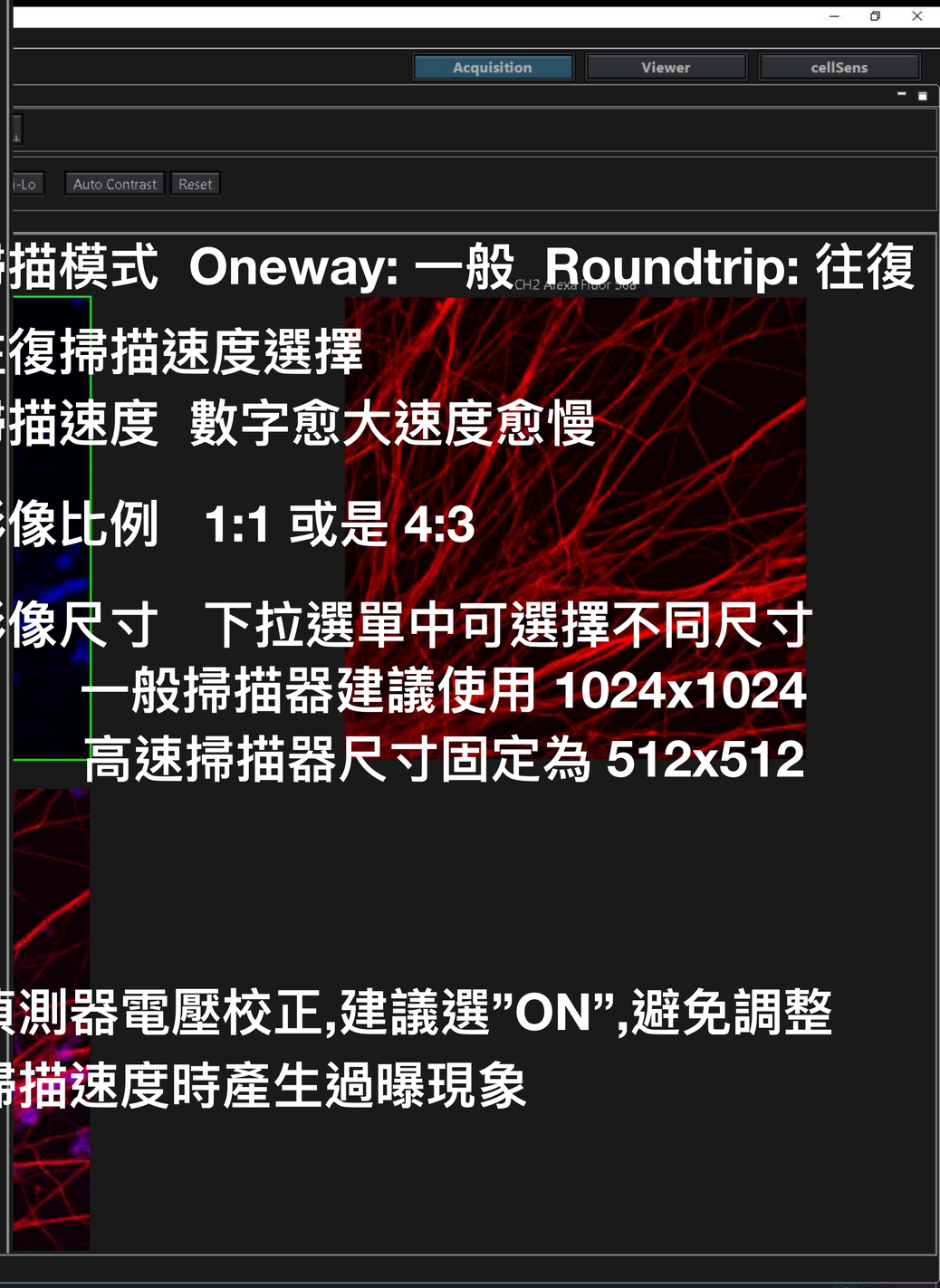


選取偵測器控制視窗中的
”Dye & Detector Select”，選取樣本
中所使用的螢光種類，按下”OK”軟體
會自動套用相對應的PMT與雷射設定

掃描器設定視窗







掃描器可自由選轉角度

光學變焦

掃描模式 Oneway: 一般 Roundtrip: 往復

往復掃描速度選擇

掃描速度 數字愈大速度愈慢

影像比例 1:1 或是 4:3

影像尺寸 下拉選單中可選擇不同尺寸

一般掃描器建議使用 1024x1024

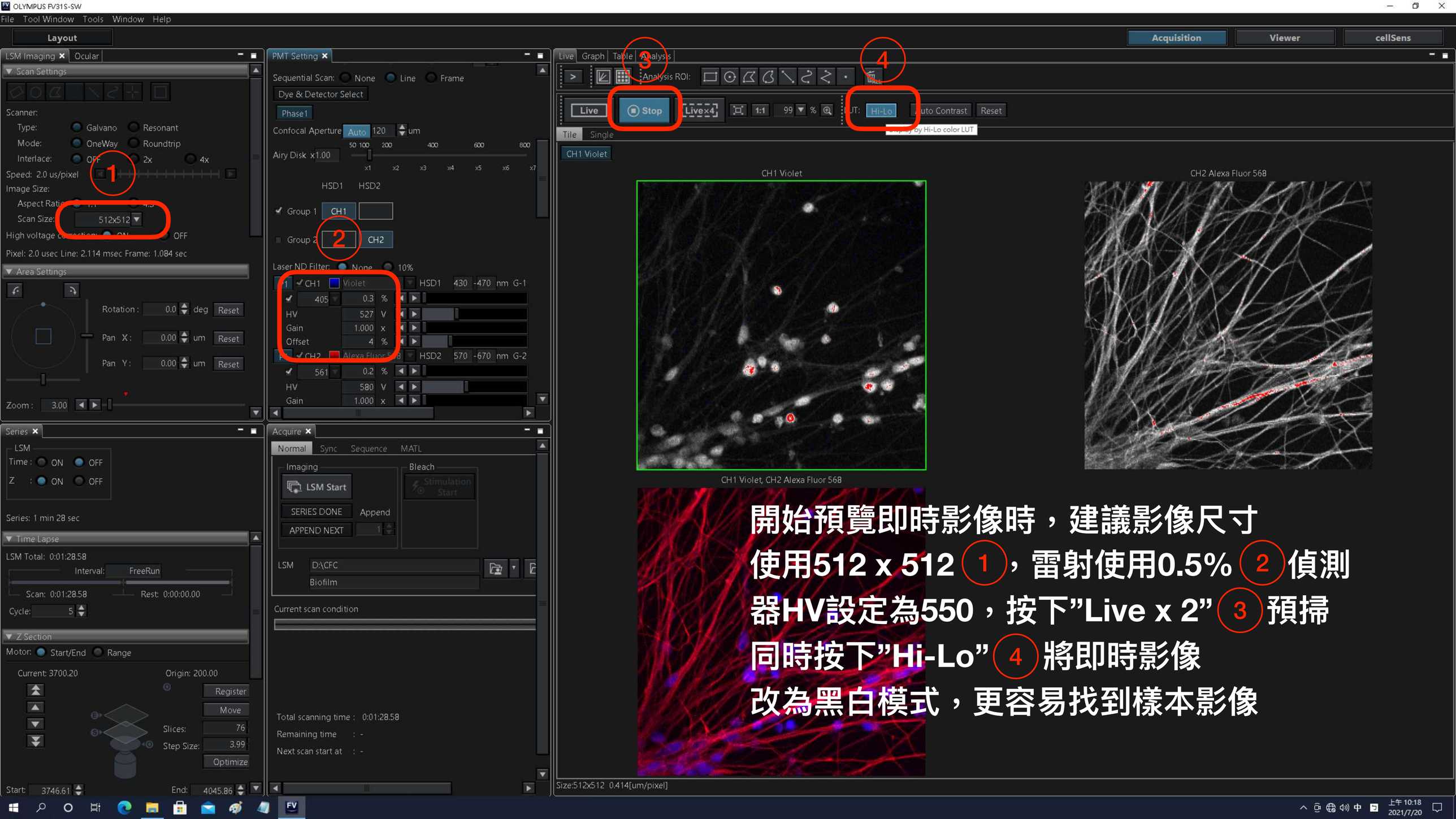
高速掃描器尺寸固定為 512x512

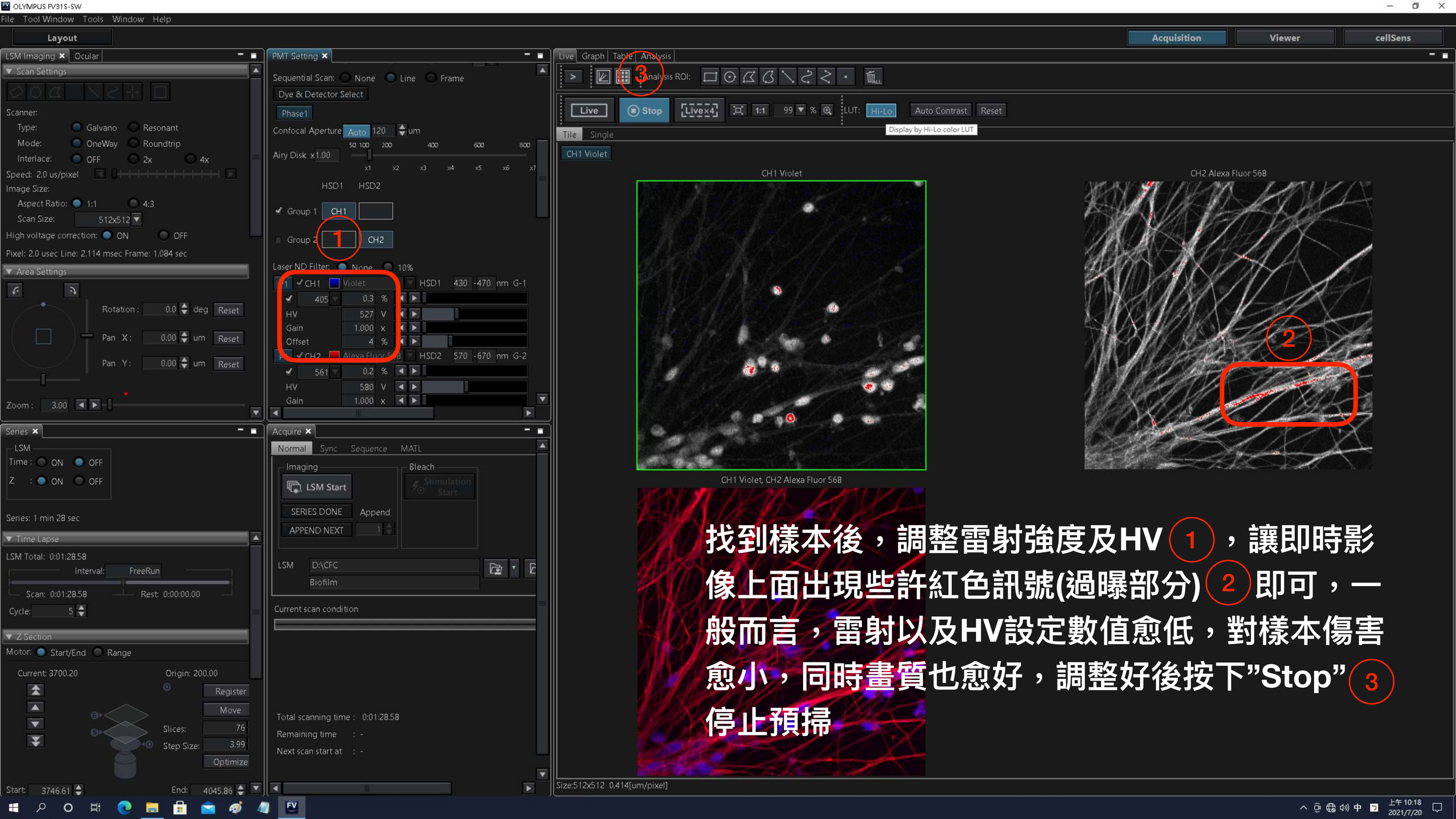
偵測器電壓校正,建議選”ON”,避免調整掃描速度時產生過曝現象

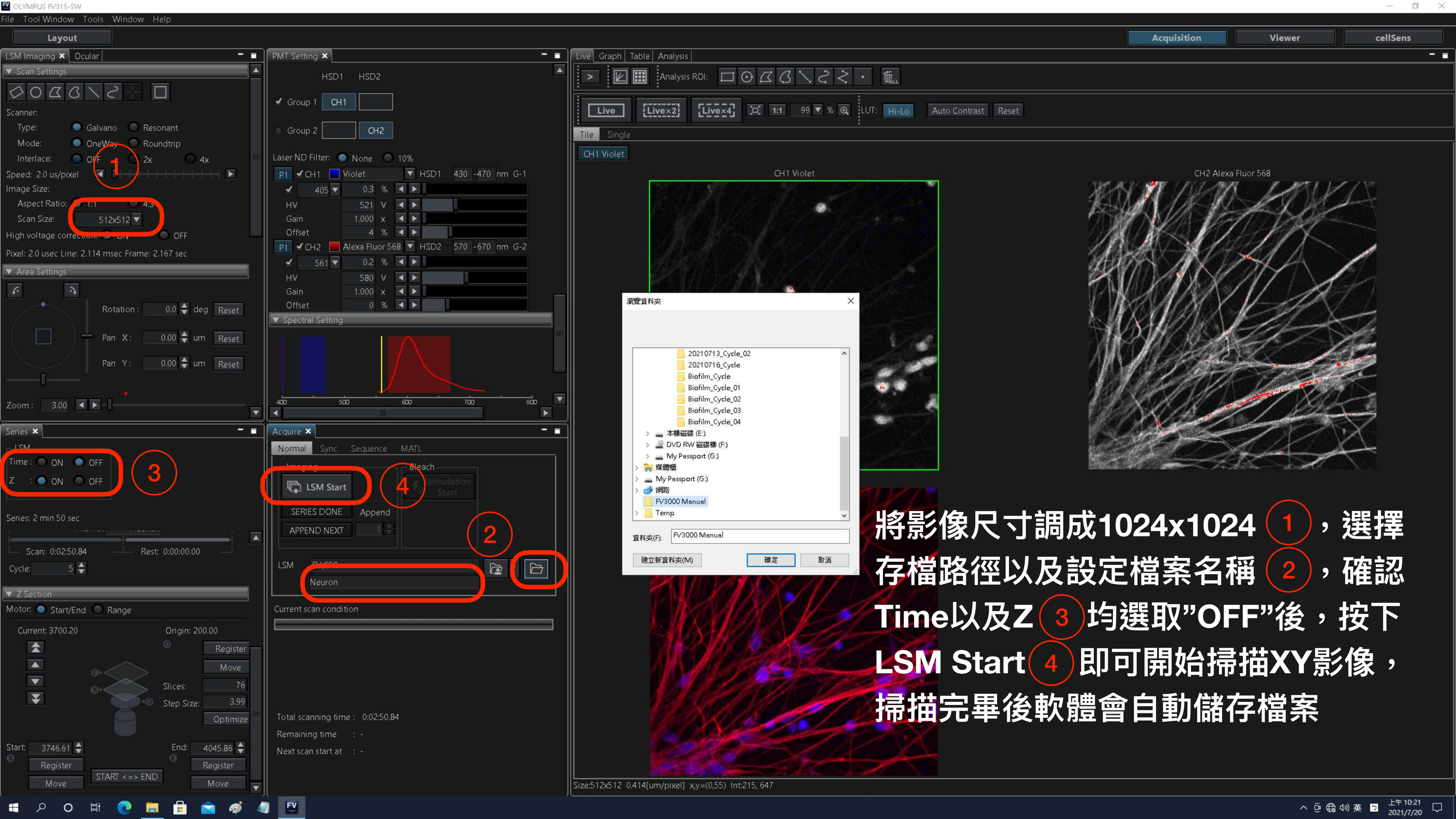
掃描器設定視窗上半部


PMT
Mode :
VBF
Lambda
偵測模式，請選VBF
Average:
None
Line
Frame
2
Times
Sequential Scan:
None
Line
Frame
影像平均，可提高S/N ratio
次數愈多效果愈好，掃描時
間相對較久
序列掃描，為避免漏光發生建議一定要開
啟，選擇Line模式可同時更新所有螢光頻
道，Frame模式一次僅更新一個頻道
Dye & Detector Select
Confocal Aperture
Auto
120
um
針孔光圈大小，請啟用Auto功能
50 100 200 400 600 800
Airy Disk x1.00
x1 x2 x3 x4 x5 x6 x7
針孔光圈大小調整，建議使用1.00
HSD1
HSD2
Group 1
CH1

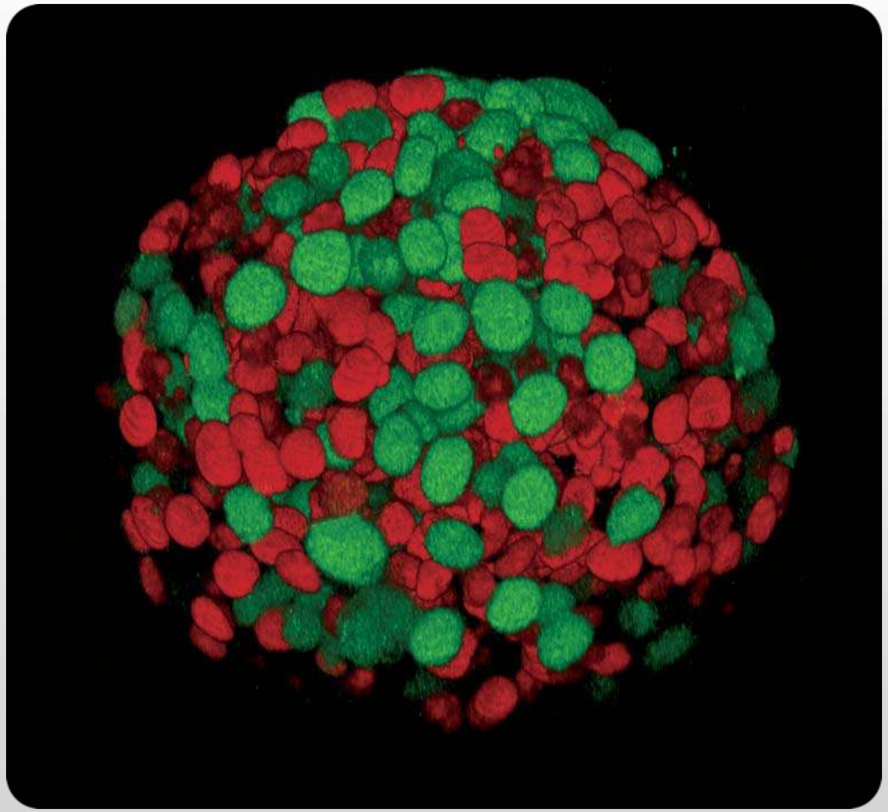

掃描器設定視窗下半部
序列掃描群組設定
雷射減光片，選取10%時，雷射最小強度可設定為0.01%
螢光種類
所使用PMT
雷射波長與強度
HV: PMT電壓,代表靈敏度,建議<750
Gain: 放大畫面上所有訊號
Offset: 過濾畫面上背景及雜訊

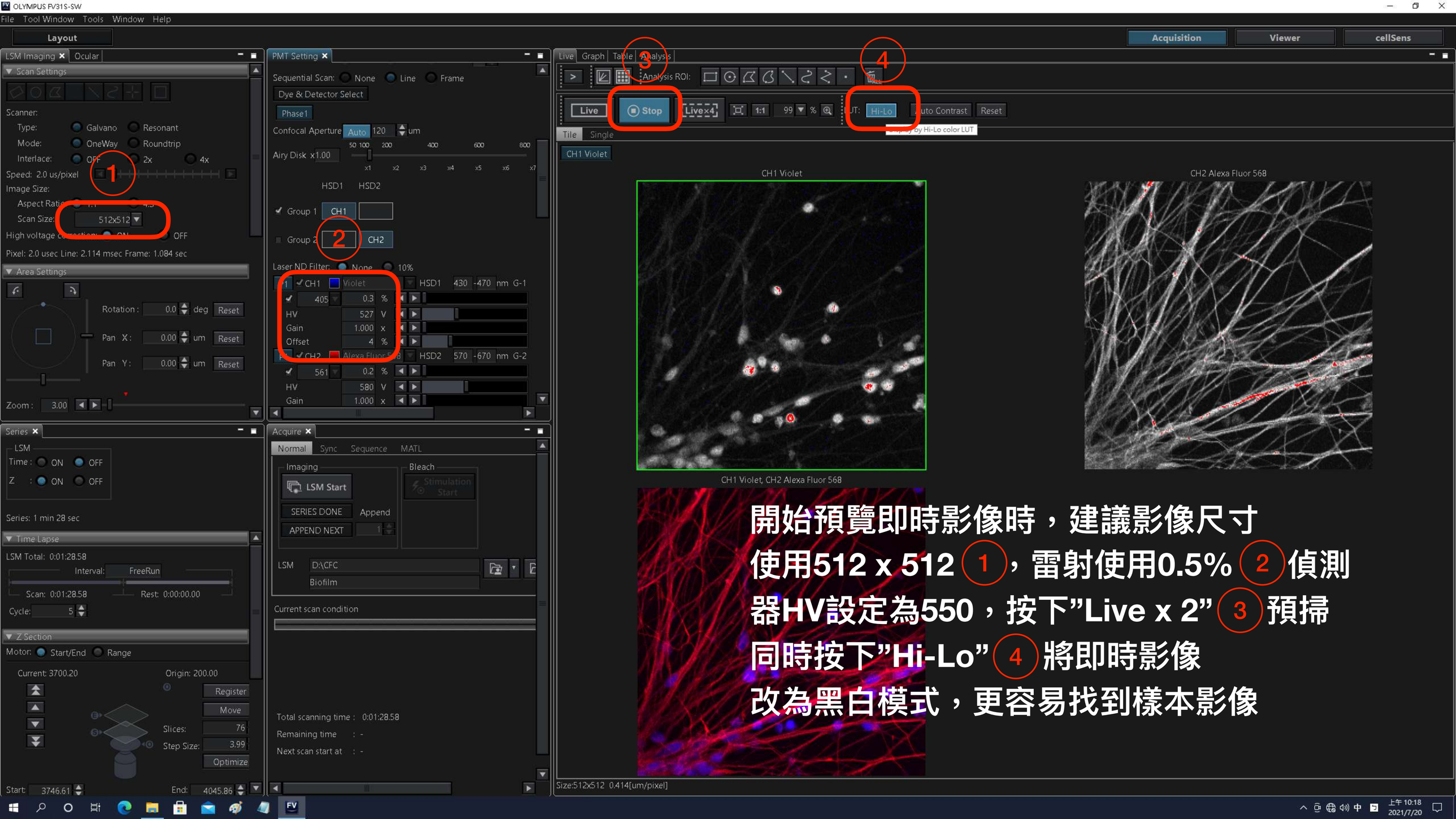

OLYMPUS FV31S-SW
File Tool Window Tools Window Help
Layout
Acquisition
Viewer
cellSens
LSM Imaging
Ocular
Scan Settings
Scanner:
Type: Galvano Resonant
Mode: OneWay Roundtrip
Interlace: OFF 2x 4x
Speed: 2.0 us/pixel
Image Size:
Aspect Ratio
Scan Size: 512x512
High voltage correction: ON OFF
Pixel: 2.0 usec Line: 2.114 msec Frame: 1.084 sec
Area Settings
Rotation: 0.0 deg Reset
Pan X: 0.00 um Reset
Pan Y: 0.00 um Reset
Zoom: 3.00
1
PMT Setting
Sequential Scan: None Line Frame
Dye & Detector Select
Phase1
Confocal Aperture Auto 120 um
Airy Disk x1.00
HSD1 HSD2
Group 1 CH1
Group 2 CH2
2
Laser ND Filter: None 10%
CH1 Violet HSD1 430 -470 nm G-1
405 0.3 %
HV 527 V
Gain 1.000 x
Offset 4 %
CH2 Alexa Fluor 568 HSD2 570 -670 nm G-2
561 0.2 %
HV 580 V
Gain 1.000 x
Series
LSM
Time: ON OFF
Z: ON OFF
Series: 1 min 28 sec
Time Lapse
LSM Total: 0:01:28.58
Interval: FreeRun
Scan: 0:01:28.58 Rest: 0:00:00.00
Cycle: 5
Z Section
Motor: Start/End Range
Current: 3700.20 Origin: 200.00
Register
Move
Slices: 76
Step Size: 3.99
Optimize
Start: 3746.61 End: 4045.86
Acquire
Normal Sync Sequence MATL
Imaging
LSM Start
SERIES DONE Append
APPEND NEXT 1
Bleach
Stimulation Start
LSM D:\CFC
Biofilm
Current scan condition
Total scanning time: 0:01:28.58
Remaining time : -
Next scan start at : -
Live Graph Table Analysis
3
4
Analysis ROI:
Live Stop Live x4 1:1 99 % LUT: Hi-Lo Auto Contrast Reset
Tile Single
CH1 Violet
CH1 Violet
CH2 Alexa Fluor 568
CH1 Violet, CH2 Alexa Fluor 568
開始預覽即時影像時，建議影像尺寸
使用512 x 512 (1)，雷射使用0.5% (2)偵測
器HV設定為550，按下”Live x 2” (3)預掃
同時按下”Hi-Lo” (4)將即時影像
改為黑白模式，更容易找到樣本影像
Size:512x512 0.414[um/pixel]
上午 10:18
2021/7/20

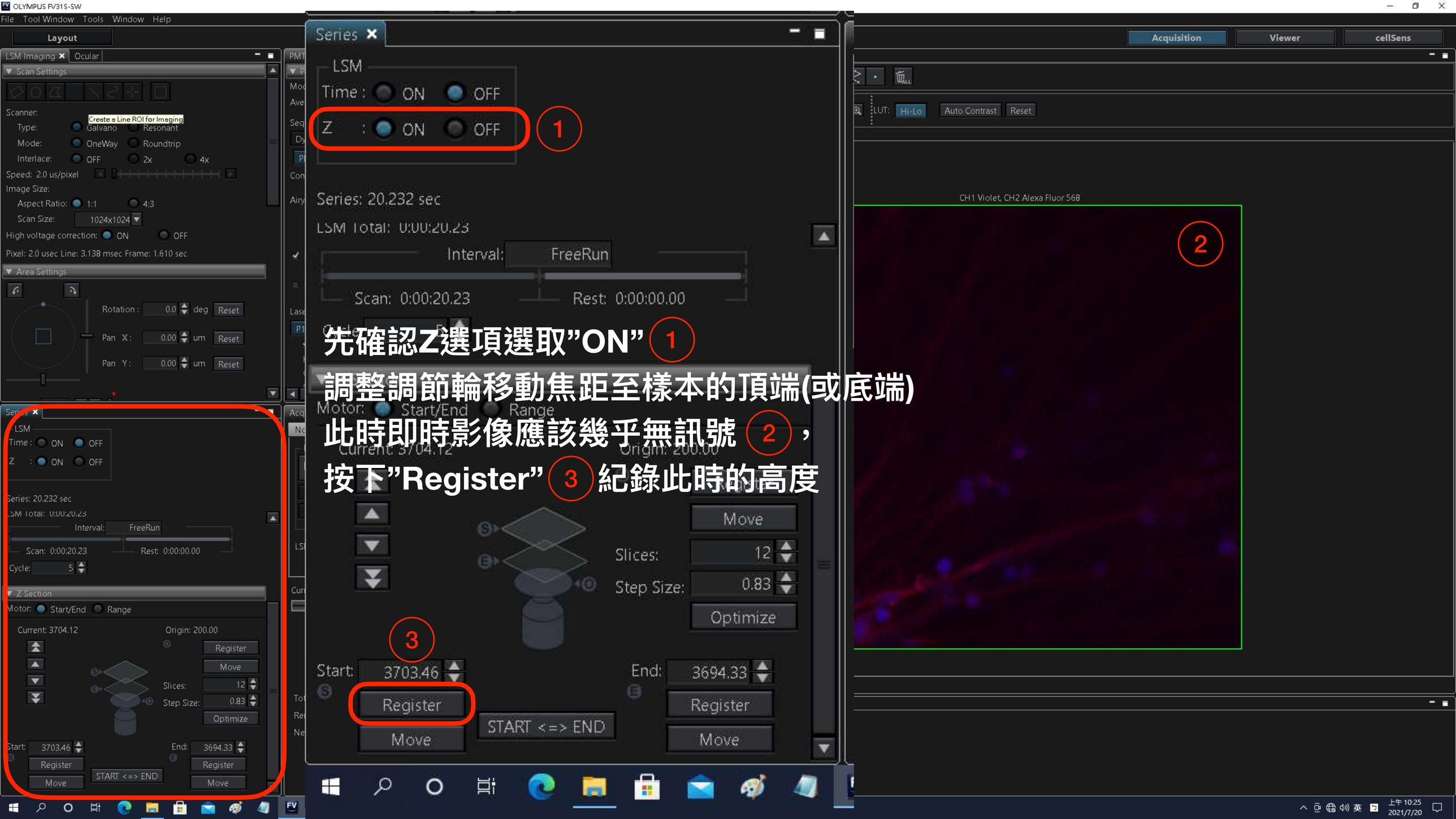

OLYMPUS FV31S-SW
File Tool Window Tools Window Help
Layout
Acquisition
Viewer
cellSens
LSM Imaging
Ocular
Scan Settings
Scanner:
Type: Galvano Resonant
Mode: OneWay Roundtrip
Interlace: OFF 2x 4x
Speed: 2.0 us/pixel
Image Size:
Aspect Ratio: 1:1 4:3
Scan Size: 512x512
High voltage correction: ON OFF
Pixel: 2.0 usec Line: 2.114 msec Frame: 1.084 sec
Area Settings
Rotation : 0.0 deg Reset
Pan X : 0.00 um Reset
Pan Y : 0.00 um Reset
Zoom : 3.00
PMT Setting
Sequential Scan: None Line Frame
Dye & Detector Select
Phase1
Confocal Aperture Auto 120 um
Airy Disk x1.00
HSD1 HSD2
Group 1 CH1
Group 2 CH2
1
Laser ND Filter: None 10%
CH1 Violet HSD1 430 -470 nm G-1
405 0.3 %
HV 527 V
Gain 1.000 x
Offset 4 %
CH2 Alexa Fluor 568 HSD2 570 -670 nm G-2
561 0.2 %
HV 580 V
Gain 1.000 x
Series
LSM
Time : ON OFF
Z : ON OFF
Series: 1 min 28 sec
Time Lapse
LSM Total: 0:01:28.58
Interval: FreeRun
Scan: 0:01:28.58 Rest: 0:00:00.00
Cycle: 5
Z Section
Motor: Start/End Range
Current: 3700.20 Origin: 200.00
Register
Move
Slices: 76
Step Size: 3.99
Optimize
Start: 3746.61 End: 4045.86
Acquire
Normal Sync Sequence MATL
Imaging
LSM Start
SERIES DONE Append
APPEND NEXT 1
Bleach
Stimulation Start
LSM D:\CFC
Biofilm
Current scan condition
Total scanning time : 0:01:28.58
Remaining time : -
Next scan start at : -
Live Graph Table Analysis
3
Analysis ROI:
Live Stop Live×4 1:1 99 % LUT: Hi-Lo Auto Contrast Reset
Display by Hi-Lo color LUT
Tile Single
CH1 Violet
CH1 Violet
CH2 Alexa Fluor 568
2
CH1 Violet, CH2 Alexa Fluor 568
找到樣本後，調整雷射強度及HV (1)，讓即時影像上面出現些許紅色訊號(過曝部分) (2) 即可，一般而言，雷射以及HV設定數值愈低，對樣本傷害愈小，同時畫質也愈好，調整好後按下”Stop” (3) 停止預掃
Size:512x512 0.414[um/pixel]
上午 10:18
2021/7/20

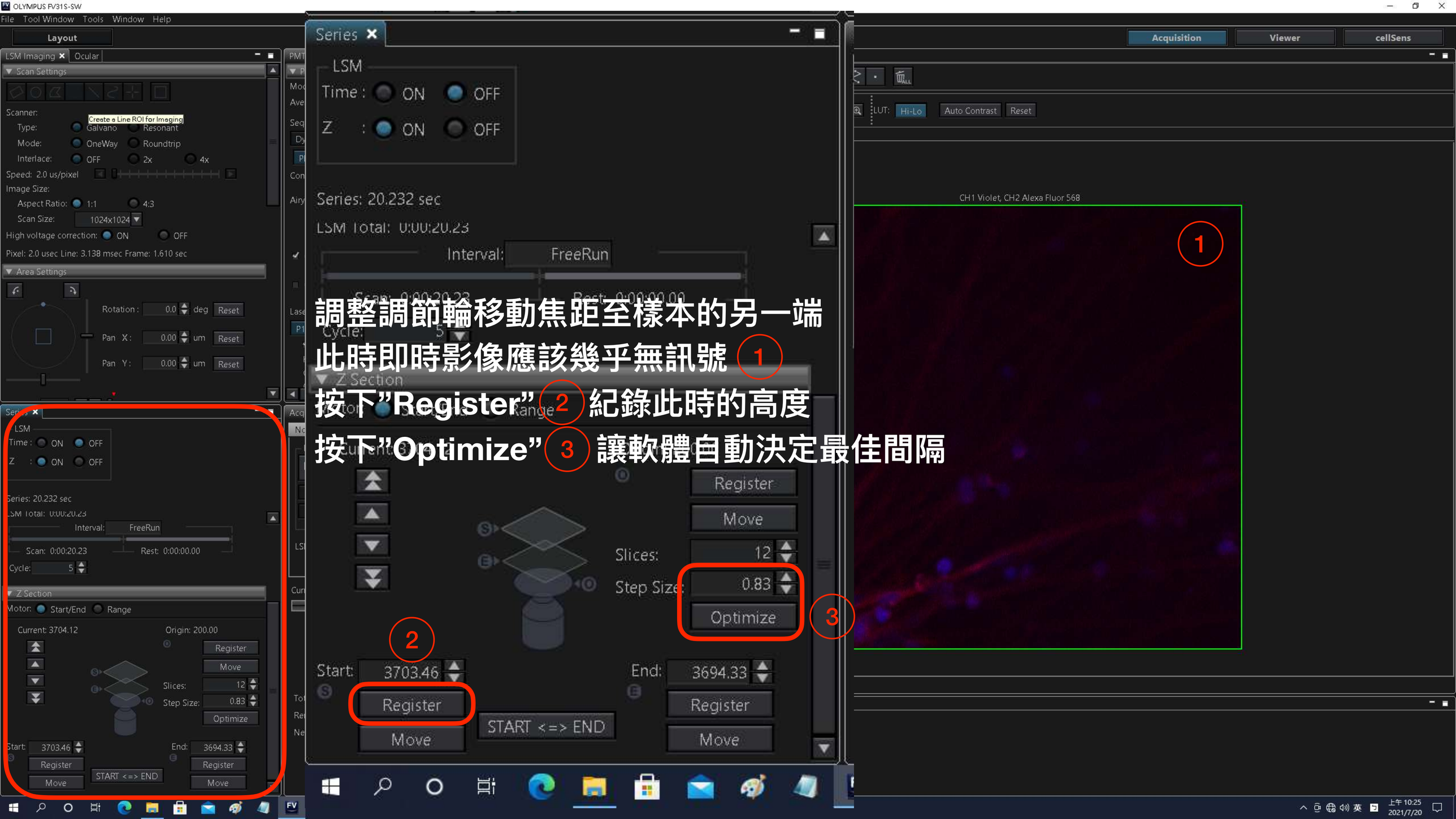

OLYMPUS FV31S-SW
File Tool Window Tools Window Help
Layout
Acquisition
Viewer
cellSens
LSM Imaging
Ocular
Scan Settings
Scanner:
Type: Galvano Resonant
Mode: OneWay Roundtrip
Interlace: OFF 2x 4x
Speed: 2.0 us/pixel
Image Size:
Aspect Ratio: 1:1 4:3
Scan Size: 512x512
High voltage correction: ON OFF
Pixel: 2.0 usec Line: 2.114 msec Frame: 2.167 sec
Area Settings
Rotation: 0.0 deg Reset
Pan X: 0.00 um Reset
Pan Y: 0.00 um Reset
Zoom: 3.00
1
PMT Setting
HSD1 HSD2
Group 1 CH1
Group 2 CH2
Laser ND Filter: None 10%
P1 CH1 Violet HSD1 430 -470 nm G-1
405 0.3 %
HV 521 V
Gain 1.000 x
Offset 4 %
P1 CH2 Alexa Fluor 568 HSD2 570 -670 nm G-2
561 0.2 %
HV 580 V
Gain 1.000 x
Offset 0 %
Spectral Setting
400 500 600 700 800
Live Graph Table Analysis
Analysis ROI:
Live Livex2 Livex4 1:1 99 % LUT: Hi-Lo Auto Contrast Reset
Tile Single
CH1 Violet
CH1 Violet
CH2 Alexa Fluor 568
瀏覽資料夾
20210713_Cycle_02
20210716_Cycle
Biofilm_Cycle
Biofilm_Cycle_01
Biofilm_Cycle_02
Biofilm_Cycle_03
Biofilm_Cycle_04
本機磁碟 (E:)
DVD RW 磁碟機 (F:)
My Passport (G:)
媒體櫃
My Passport (G:)
網路
FV3000 Manual
Temp
資料夾(F): FV3000 Manual
建立新資料夾(M)
確定
取消
Series
LSM
Time: ON OFF
Z: ON OFF
3
Series: 2 min 50 sec
Scan: 0:02:50.84
Rest: 0:00:00.00
Cycle: 5
Z Section
Motor: Start/End Range
Current: 3700.20
Origin: 200.00
Register
Move
Slices: 76
Step Size: 3.99
Optimize
Start: 3746.61
End: 4045.86
Register
Move
START <=> END
Acquire
Normal Sync Sequence MATL
Bleach
LSM Start
4
Stimulation Start
SERIES DONE
Append
APPEND NEXT
1
2
LSM
Neuron
Current scan condition
Total scanning time: 0:02:50.84
Remaining time: -
Next scan start at: -
Size:512x512 0.414[um/pixel] x,y=(0,55) Int:215, 647
將影像尺寸調成1024x1024 1，選擇
存檔路徑以及設定檔案名稱 2，確認
Time以及Z 3 均選取”OFF”後，按下
LSM Start 4 即可開始掃描XY影像，
掃描完畢後軟體會自動儲存檔案
上午 10:21
2021/7/20

XYZ(Z stack)取像 Step by step




OLYMPUS FV31S-SW
File Tool Window Tools Window Help
Layout
Acquisition
Viewer
cellSens
LSM Imaging
Ocular
Scan Settings
Scanner:
Type: Galvano Resonant
Mode: OneWay Roundtrip
Interlace: OFF 2x 4x
Speed: 2.0 us/pixel
Image Size:
Aspect Ratio
Scan Size: 512x512
High voltage correction: ON OFF
Pixel: 2.0 usec Line: 2.114 msec Frame: 1.084 sec
Area Settings
Rotation: 0.0 deg Reset
Pan X: 0.00 um Reset
Pan Y: 0.00 um Reset
Zoom: 3.00
1
PMT Setting
Sequential Scan: None Line Frame
Dye & Detector Select
Phase1
Confocal Aperture Auto 120 um
Airy Disk x1.00
HSD1 HSD2
Group 1 CH1
Group 2 CH2
2
Laser ND Filter: None 10%
CH1 Violet HSD1 430 -470 nm G-1
405 0.3 %
HV 527 V
Gain 1.000 x
Offset 4 %
CH2 Alexa Fluor 568 HSD2 570 -670 nm G-2
561 0.2 %
HV 580 V
Gain 1.000 x
Series
LSM
Time: ON OFF
Z: ON OFF
Series: 1 min 28 sec
Time Lapse
LSM Total: 0:01:28.58
Interval: FreeRun
Scan: 0:01:28.58 Rest: 0:00:00.00
Cycle: 5
Z Section
Motor: Start/End Range
Current: 3700.20 Origin: 200.00
Register
Move
Slices: 76
Step Size: 3.99
Optimize
Start: 3746.61 End: 4045.86
Acquire
Normal Sync Sequence MATL
Imaging
LSM Start
SERIES DONE Append
APPEND NEXT 1
Bleach
Stimulation Start
LSM D:\CFC
Biofilm
Current scan condition
Total scanning time: 0:01:28.58
Remaining time : -
Next scan start at : -
Live Graph Table Analysis
3
4
Analysis ROI:
Live Stop Live x4 1:1 99 % LUT: Hi-Lo Auto Contrast Reset
Tile Single
CH1 Violet
CH1 Violet
CH2 Alexa Fluor 568
CH1 Violet, CH2 Alexa Fluor 568
開始預覽即時影像時，建議影像尺寸
使用512 x 512 (1)，雷射使用0.5% (2)偵測
器HV設定為550，按下”Live x 2” (3)預掃
同時按下”Hi-Lo” (4)將即時影像
改為黑白模式，更容易找到樣本影像
Size:512x512 0.414[um/pixel]
上午 10:18
2021/7/20


Series
LSM
Time : ON OFF
Z : ON OFF
1
Series: 20.232 sec
LSM Total: 0:00:20.23
Interval: FreeRun
Scan: 0:00:20.23
Rest: 0:00:00.00
先確認Z選項選取”ON” 1
調整調節輪移動焦距至樣本的頂端(或底端)
此時即時影像應該幾乎無訊號 2 ，
按下”Register” 3 紀錄此時的高度
Motor: Start/End Range
Move
Slices: 12
Step Size: 0.83
Optimize
3
Start: 3703.46
Register
Move
START <=> END
End: 3694.33
Register
Move
2
CH1 Violet, CH2 Alexa Fluor 568


調整調節輪移動焦距至樣本的另一端
此時即時影像應該幾乎無訊號 1
按下”Register” 2 紀錄此時的高度
按下”Optimize” 3 讓軟體自動決定最佳間隔
Series
LSM
Time : ON OFF
Z : ON OFF
Series: 20.232 sec
Interval: FreeRun
Z Section
Register
Move
Slices: 12
Step Size: 0.83
Optimize
Start: 3703.46
End: 3694.33
START <=> END
CH1 Violet, CH2 Alexa Fluor 568

OLYMPUS FV31S-SW

File Tool Window Tools Window Help

Layout | Acquisition | Viewer | cellSens

LSM Imaging | Ocular

Scan Settings

Scanner:
Type: Galvano Resonant
Mode: OneWay Roundtrip
Interlace: OFF 2x 4x
Speed: 2.0 us/pixel
Image Size:
Aspect Ratio: 1:1 4:3
Scan Size: 800x800
High voltage correction: ON OFF
Pixel: 2.0 usec Line: 2.690 msec Frame: 8.611 sec

Area Settings

Rotation: 0.0 deg Reset
Pan X: 0.00 um Reset
Pan Y: 0.00 um Reset

PMT Setting

PMT

Mode: VBF Lambda
Average: None Line Frame 2 Times
Sequential Scan: None Line Frame
Dye & Detector Select
Phase1
Confocal Aperture: Auto 120 um
50 100 200 400 600 800
Airy Disk x1.00
x1 x2 x3 x4 x5 x6 x7
HSD1 HSD2
Group 1 CH1
Group 2 CH2
Laser ND Filter: None 10%
P1 CH1 Violet HSD1 430 -470 nm G-1
405 0.3 %
HV 521 V
Gain 1.000 x
Offset 4 %

Series

LSM
Time: ON OFF
Z: ON OFF
Series: 1 min 44 sec
LSM Total: 0:01:44.24
Interval: FreeRun
Scan: 0:01:44.24 Rest: 0:00:00.00
Cycle: 5

Z Section

Motor: Start/End Range
Current: 3702.63 Origin: 200.00
Register
Move
Slices: 12
Step Size: 0.83
Optimize
Start: 3703.46 Register Move
START <=> END
End: 3694.33 Register Move

*Acquire

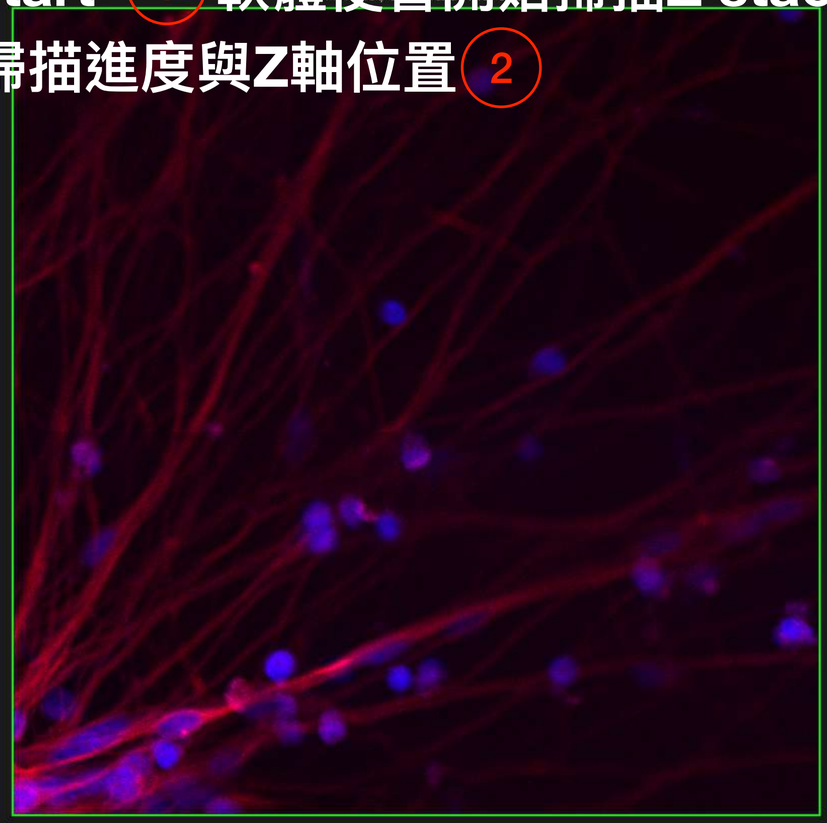
Normal Sync Sequence MATL
Imaging
LSM Stop
LSM scan
APPEND NEXT 1
Bleach
Stimulation Start
LSM D:\CFC
Neuron
Current scan condition
Z position : 1 / 12
Total scanning time : 0:01:44.24
Remaining time : 0:01:31
Next scan start at : -

1
2

Live Graph Table Analysis Neuron Neuron_0001

Analysis ROI:
Live Live x2 Live x4 1:1 100 % LUT: Hi-Lo Auto Contrast Reset
Tile Single
CH1 Violet CH2 Alexa Fluor...

按下”LSM Start” 1 軟體便會開始掃描Z stack影像
此處會顯示掃描進度與Z軸位置 2

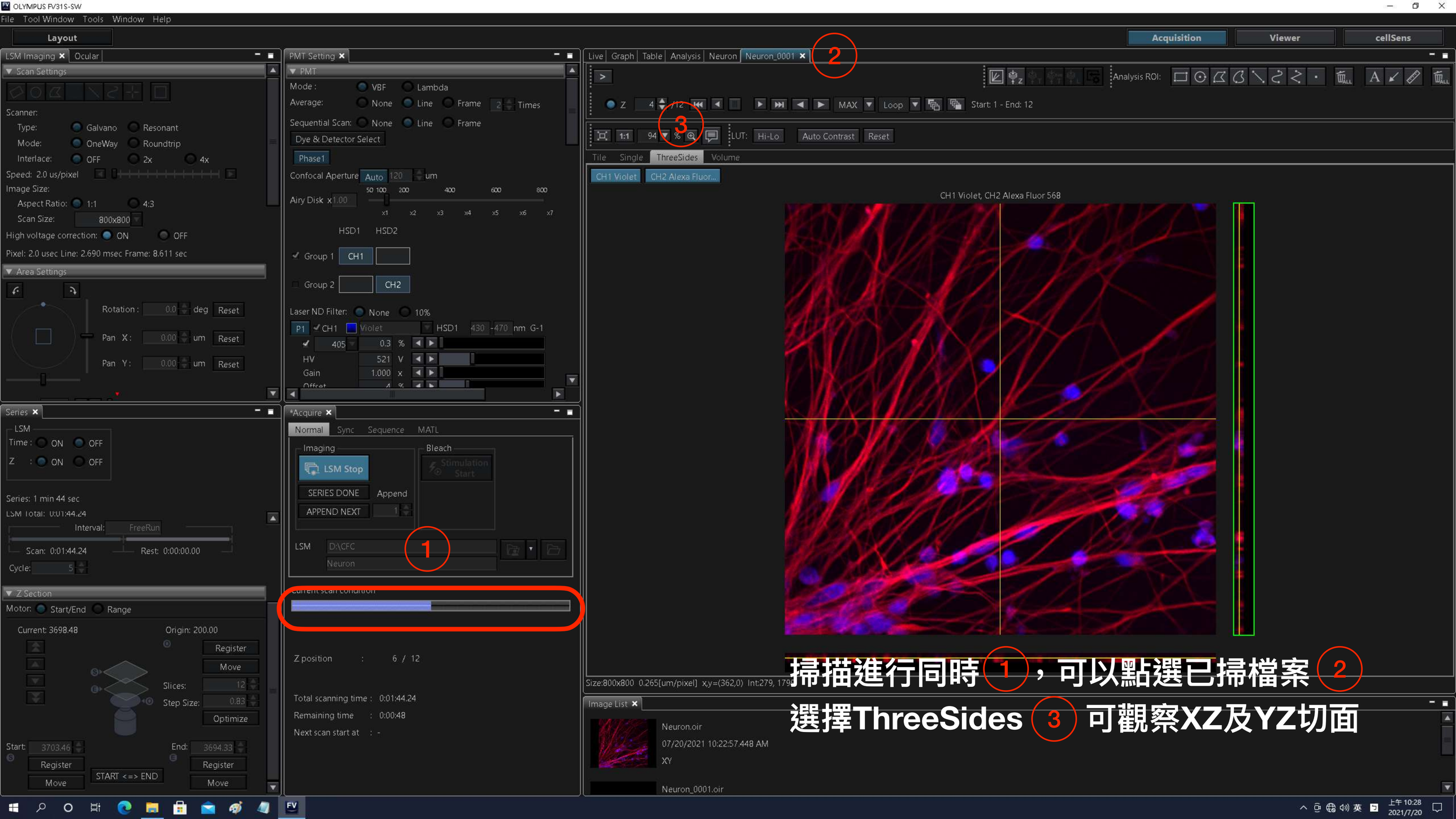


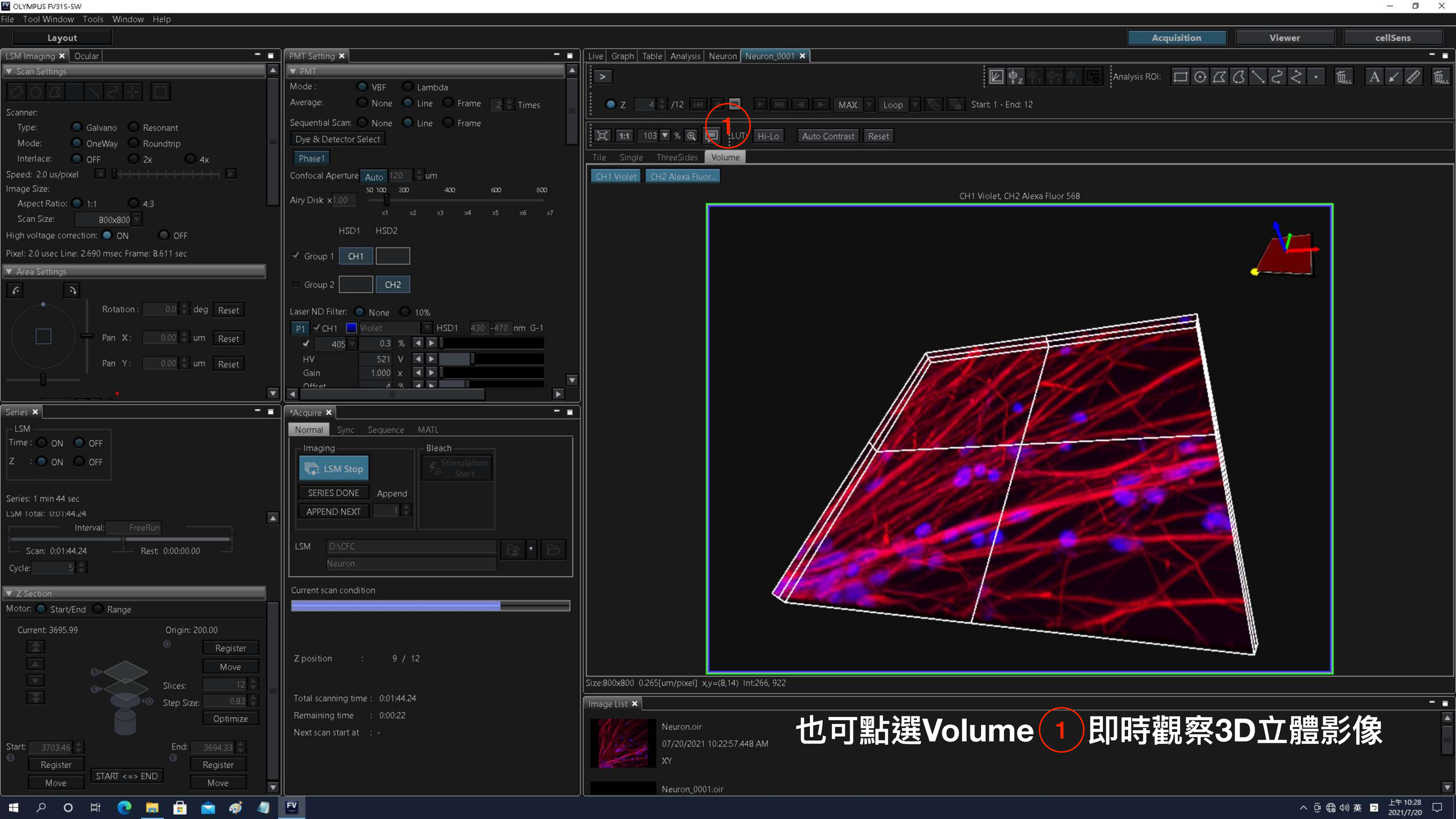
Size:800x800 0.265[um/pixel]

Image List

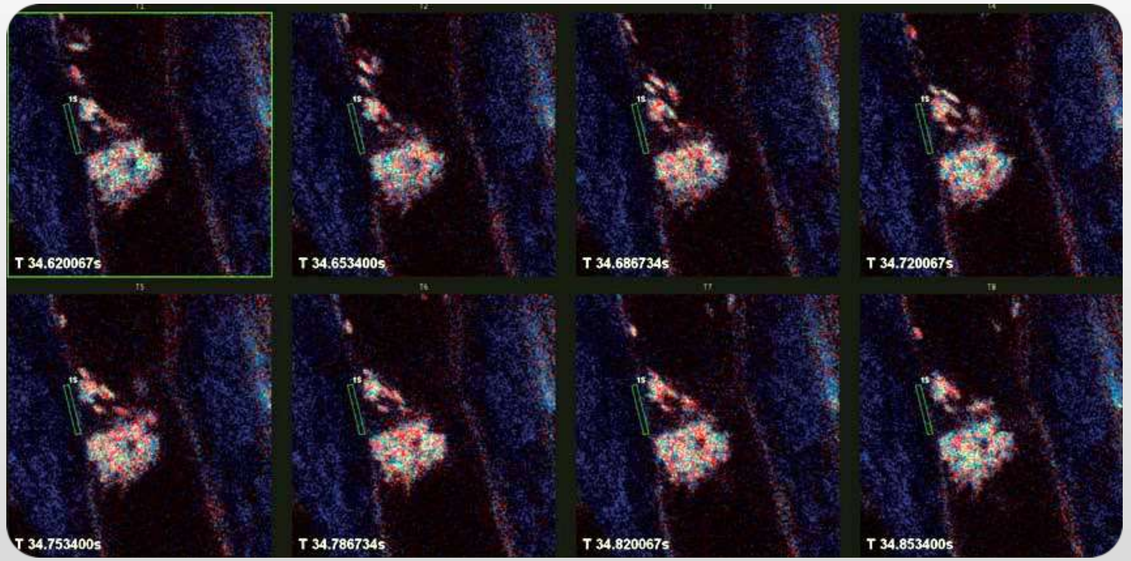
Neuron.oir
07/20/2021 10:22:57.448 AM
XY
Neuron_0001.oir

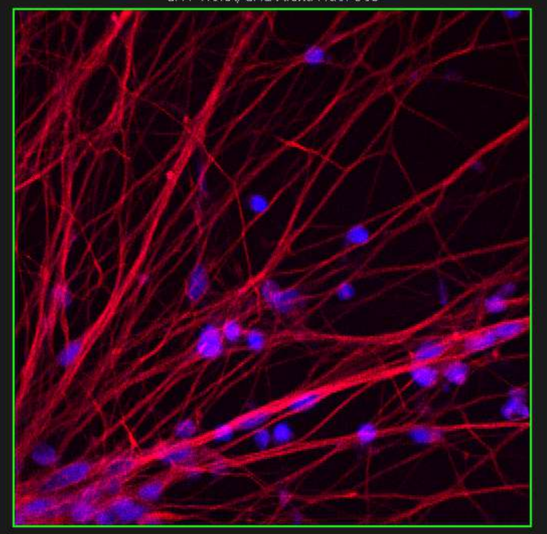
上午 10:27
2021/7/20


OLYMPUS FV31S-SW
File Tool Window Tools Window Help
Layout
Acquisition
Viewer
cellSens
LSM Imaging
Ocular
Scan Settings
Scanner:
Type: Galvano Resonant
Mode: OneWay Roundtrip
Interlace: OFF 2x 4x
Speed: 2.0 us/pixel
Image Size:
Aspect Ratio: 1:1 4:3
Scan Size: 800x800
High voltage correction: ON OFF
Pixel: 2.0 usec Line: 2.690 msec Frame: 8.611 sec
Area Settings
Rotation : 0.0 deg Reset
Pan X : 0.00 um Reset
Pan Y : 0.00 um Reset
PMT Setting
PMT
Mode : VBF Lambda
Average: None Line Frame 2 Times
Sequential Scan: None Line Frame
Dye & Detector Select
Phase1
Confocal Aperture Auto 120 um
Airy Disk x1.00
HSD1 HSD2
Group 1 CH1
Group 2 CH2
Laser ND Filter: None 10%
P1 CH1 Violet HSD1 430 -470 nm G-1
405 0.3 %
HV 521 V
Gain 1.000 x
Series
LSM
Time : ON OFF
Z : ON OFF
Series: 1 min 44 sec
LSM Total: 0:01:44.24
Interval: FreeRun
Scan: 0:01:44.24
Rest: 0:00:00.00
Cycle: 5
Z Section
Motor: Start/End Range
Current: 3698.48
Origin: 200.00
Register
Move
Slices: 12
Step Size: 0.83
Optimize
Start: 3703.46
End: 3694.33
START <=> END
*Acquire
Normal Sync Sequence MATL
Imaging
LSM Stop
SERIES DONE
Append
APPEND NEXT
Bleach
Stimulation Start
LSM D:\CFC
Neuron
1
Current scan condition
Z position : 6 / 12
Total scanning time : 0:01:44.24
Remaining time : 0:00:48
Next scan start at : -
Live Graph Table Analysis Neuron Neuron_0001
2
Analysis ROI:
Z 4 /12 MAX Loop Start: 1 - End: 12
3
1:1 94 % LUT: Hi-Lo Auto Contrast Reset
Tile Single ThreeSides Volume
CH1 Violet CH2 Alexa Fluor...
CH1 Violet, CH2 Alexa Fluor 568
Size:800x800 0.265[um/pixel] x,y=(362,0) Int:279, 179
Image List
Neuron.oir
07/20/2021 10:22:57.448 AM
XY
Neuron_0001.oir
掃描進行同時 1 ，可以點選已掃檔案 2
選擇ThreeSides 3 可觀察XZ及YZ切面


OLYMPUS FV31S-SW
File Tool Window Tools Window Help
Layout
Acquisition
Viewer
cellSens
LSM Imaging
Ocular
Scan Settings
Scanner:
Type: Galvano Resonant
Mode: OneWay Roundtrip
Interlace: OFF 2x 4x
Speed: 2.0 us/pixel
Image Size:
Aspect Ratio: 1:1 4:3
Scan Size: 800x800
High voltage correction: ON OFF
Pixel: 2.0 usec Line: 2.690 msec Frame: 8.611 sec
Area Settings
Rotation : 0.0 deg Reset
Pan X : 0.00 um Reset
Pan Y : 0.00 um Reset
PMT Setting
PMT
Mode : VBF Lambda
Average: None Line Frame 2 Times
Sequential Scan: None Line Frame
Dye & Detector Select
Phase1
Confocal Aperture Auto 120 um
Airy Disk x1.00
HSD1 HSD2
Group 1 CH1
Group 2 CH2
Laser ND Filter: None 10%
P1 CH1 Violet HSD1 430 -470 nm G-1
405 0.3 %
HV 521 V
Gain 1.000 x
Series
LSM
Time : ON OFF
Z : ON OFF
Series: 1 min 44 sec
LSM Total: 0:01:44.24
Interval: FreeRun
Scan: 0:01:44.24 Rest: 0:00:00.00
Cycle: 5
Z Section
Motor: Start/End Range
Current: 3695.99
Origin: 200.00
Register
Move
Slices: 12
Step Size: 0.83
Optimize
Start: 3703.46
End: 3694.33
START <=> END
*Acquire
Normal Sync Sequence MATL
Imaging
LSM Stop
SERIES DONE
Append
APPEND NEXT
Bleach
Stimulation Start
LSM D:\CFC
Neuron
Current scan condition
Z position : 9 / 12
Total scanning time : 0:01:44.24
Remaining time : 0:00:22
Next scan start at : -
Live Graph Table Analysis Neuron Neuron_0001
Analysis ROI:
Z 4 /12
MAX Loop Start: 1 - End: 12
1:1 103 % LUT Hi-Lo Auto Contrast Reset
1
Tile Single ThreeSides Volume
CH1 Violet CH2 Alexa Fluor...
CH1 Violet, CH2 Alexa Fluor 568
Size:800x800 0.265[um/pixel] x,y=(8,14) Int:266, 922
Image List
Neuron.oir
07/20/2021 10:22:57.448 AM
XY
Neuron_0001.oir
也可點選Volume 1 即時觀察3D立體影像

XYT(Time Lapse)取像 Step by step





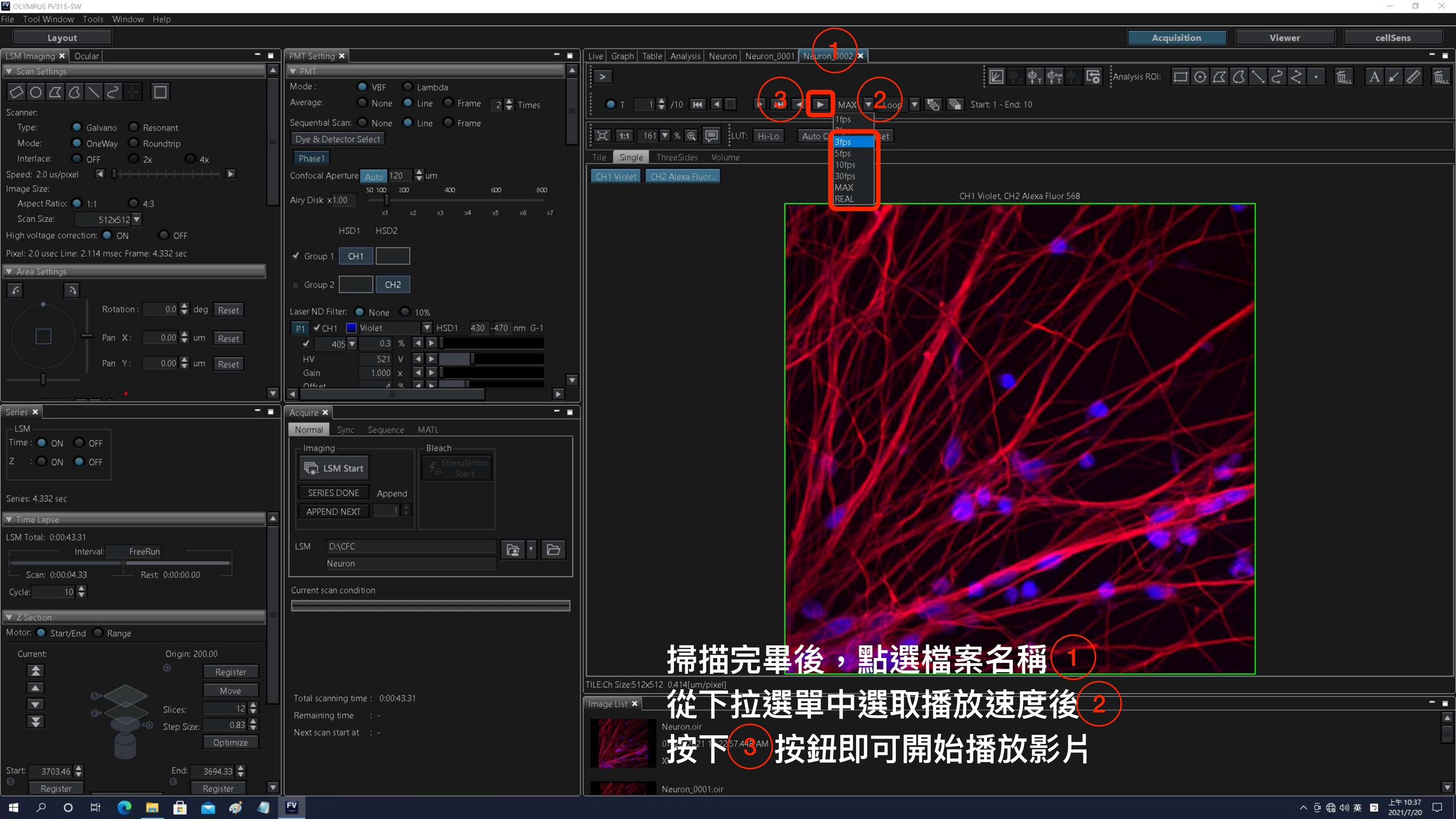
先確認Time選項選取”ON”(1)

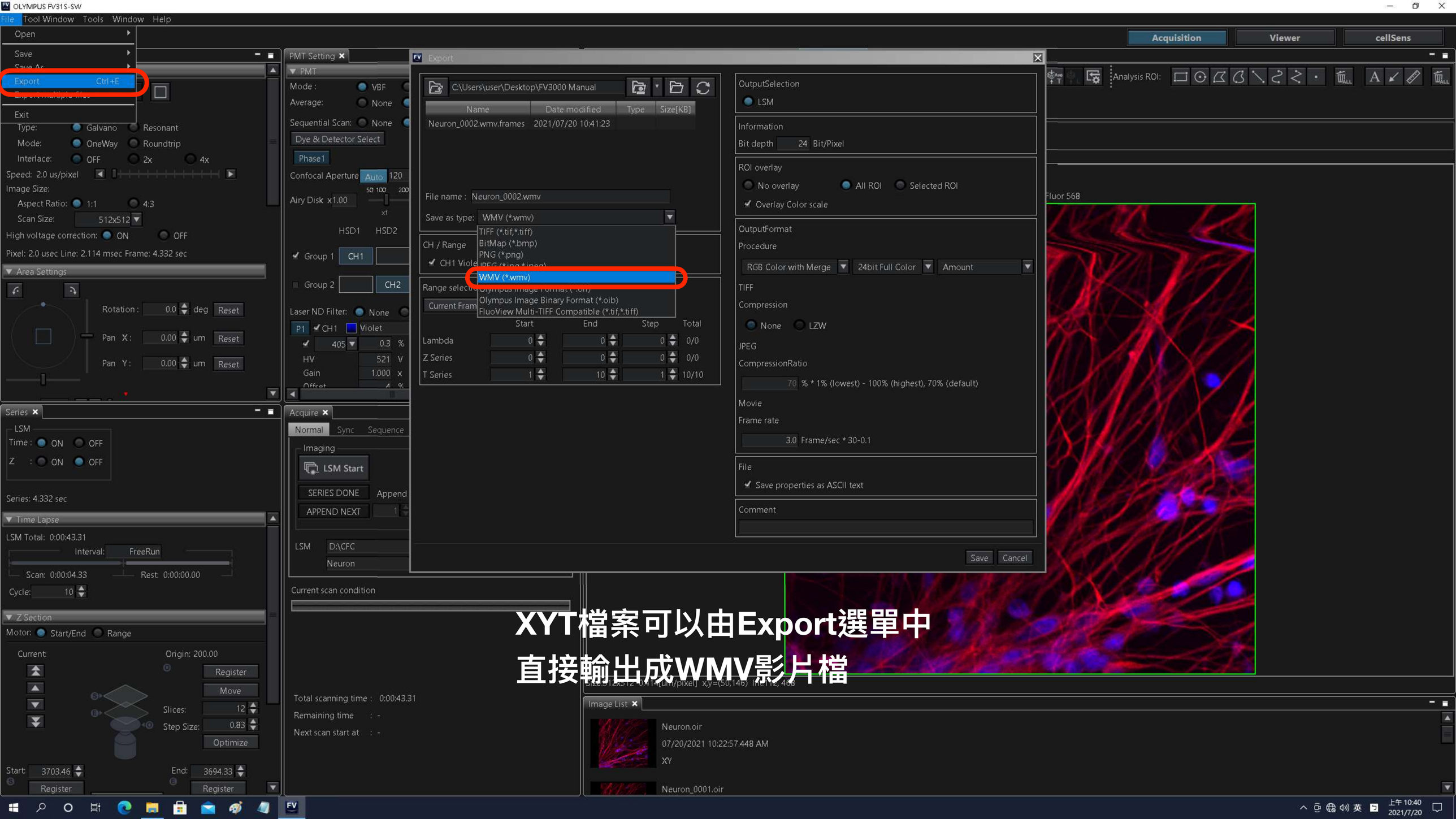
Interval(2)設定拍攝間隔，以秒為單位

例如想要五分鐘拍一張，請輸入”300”

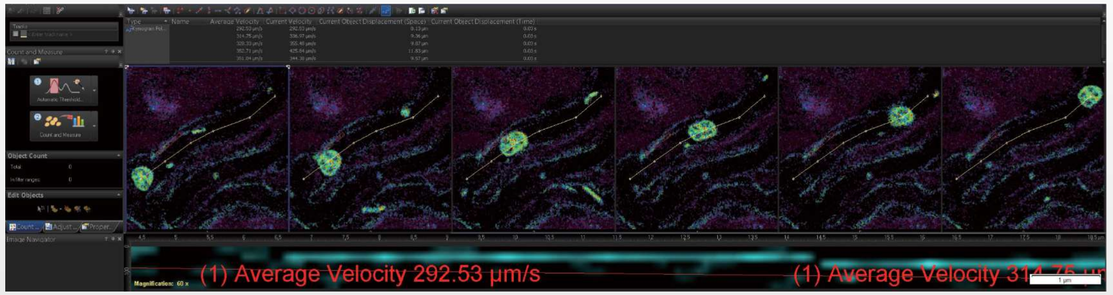
Cycle(3)設定拍攝循環數，輸入數值後

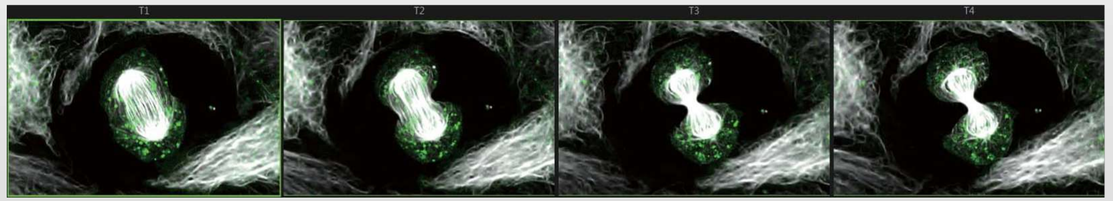
這邊會顯示總拍攝時間(4)

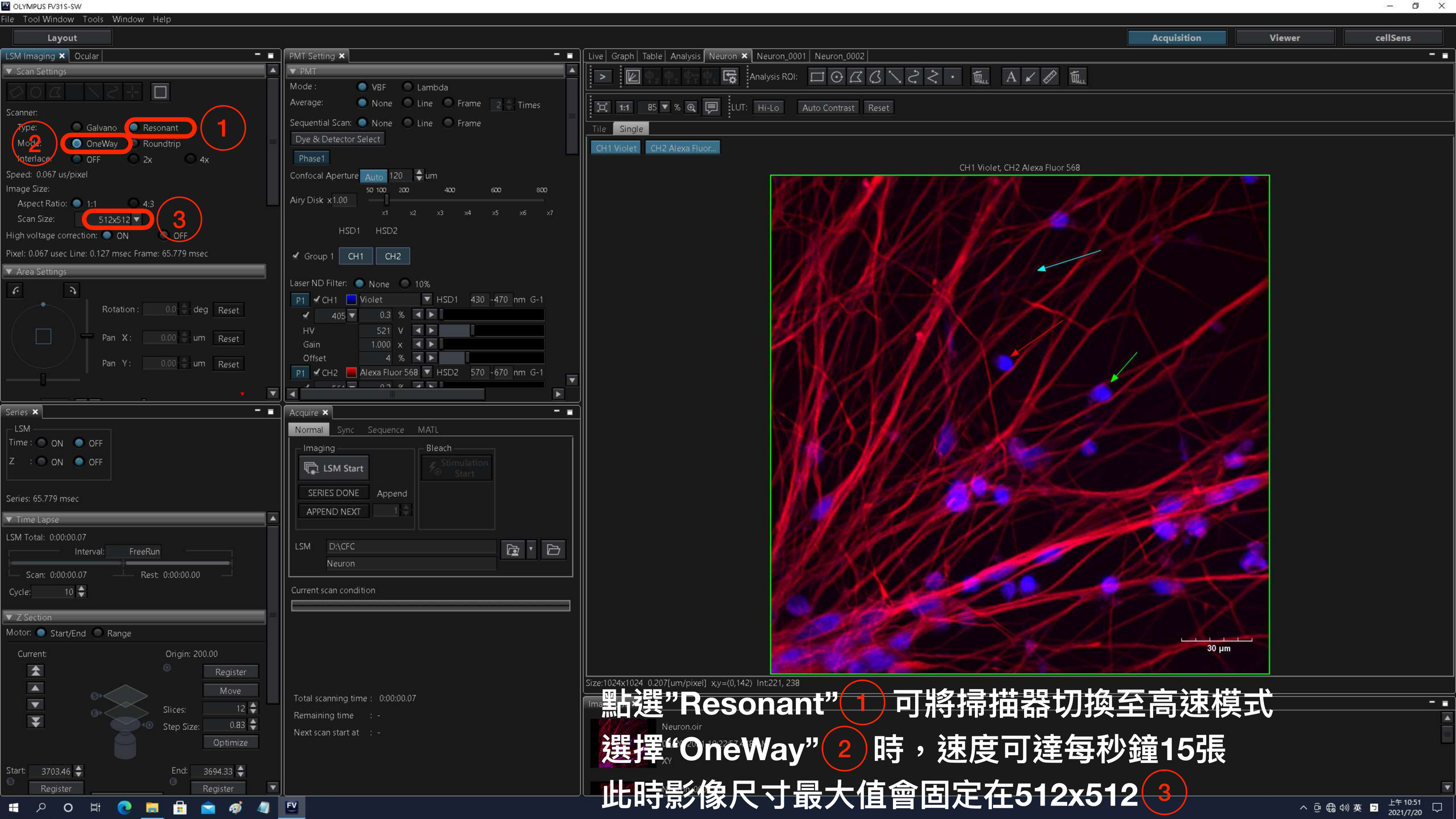

掃描完畢後，點選檔案名稱 1
從下拉選單中選取播放速度後 2
按下 3 按鈕即可開始播放影片

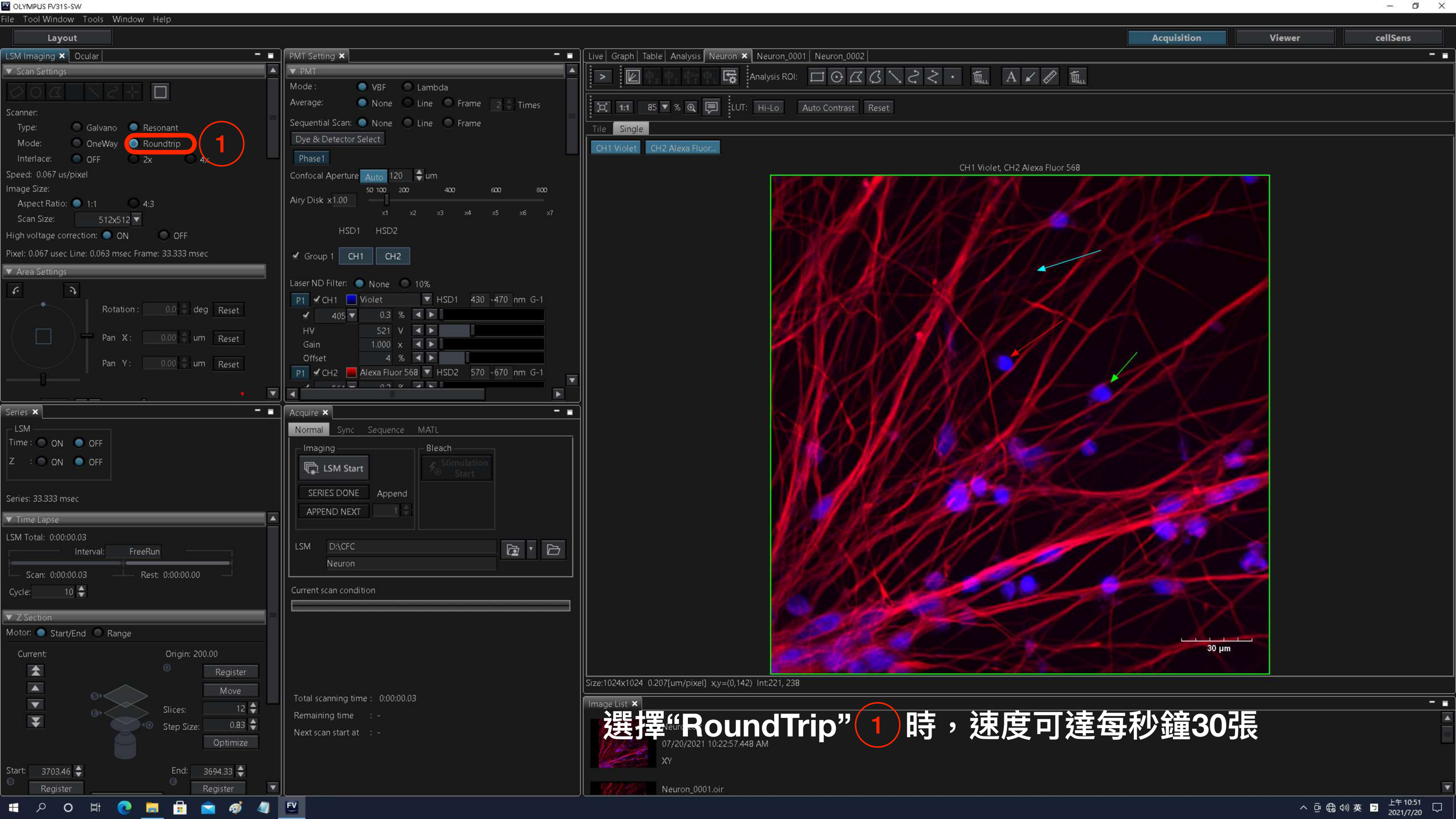

OLYMPUS FV31S-SW
File Tool Window Tools Window Help
Open
Save
Export Ctrl+E
Exit
Acquisition
Viewer
cellSens
PMT Setting
PMT
Export
C:\Users\user\Desktop\FV3000 Manual
Name
Date modified
Type
Size[KB]
Neuron_0002.wmv.frames
2021/07/20 10:41:23
File name : Neuron_0002.wmv
Save as type: WMV (*.wmv)
TIFF (*.tif,*.tiff)
BitMap (*.bmp)
PNG (*.png)
WMV (*.wmv)
Olympus Image Binary Format (*.oib)
FluoView Multi-TIFF Compatible (*.tif,*.tiff)
OutputSelection
LSM
Information
Bit depth 24 Bit/Pixel
ROI overlay
No overlay
All ROI
Selected ROI
Overlay Color scale
OutputFormat
Procedure
RGB Color with Merge
24bit Full Color
Amount
TIFF
Compression
None
LZW
JPEG
CompressionRatio
% * 1% (lowest) - 100% (highest), 70% (default)
Movie
Frame rate
3.0 Frame/sec * 30-0.1
File
Save properties as ASCII text
Comment
Save
Cancel
Start
End
Step
Total
Lambda
Z Series
T Series
Fluor 568
Analysis ROI:
Series
Time Lapse
Z Section
Acquire
Normal
Sync
Sequence
LSM Start
SERIES DONE
APPEND NEXT
Current scan condition
Total scanning time : 0:00:43.31
Remaining time : -
Next scan start at : -
Image List
Neuron.oir
07/20/2021 10:22:57.448 AM
XY
Neuron_0001.oir
XYT檔案可以由Export選單中
直接輸出成WMV影片檔

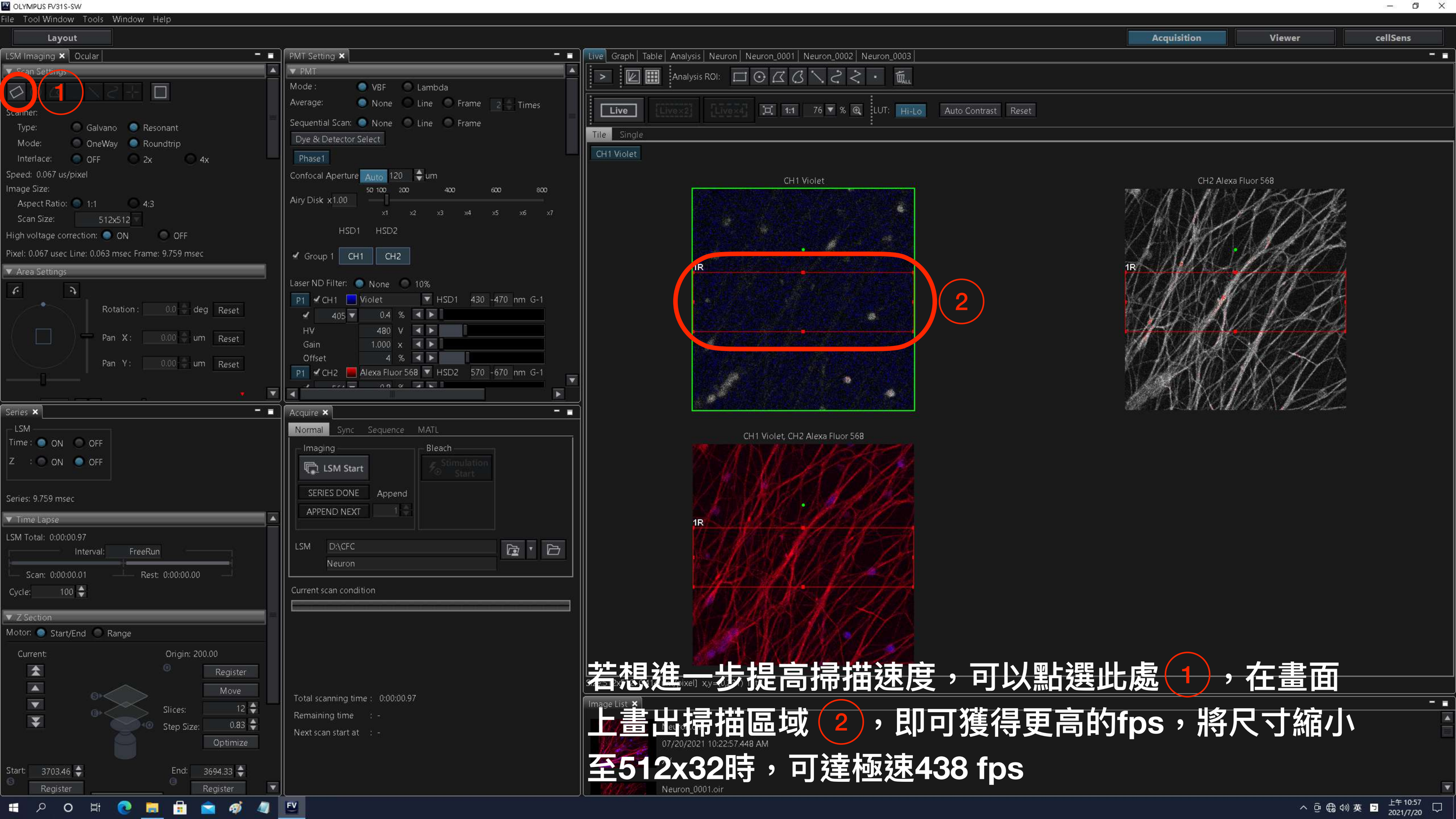
共振式掃描器(Resonant scanner)
XYT(Time Lapse)取像 Step by step






OLYMPUS FV31S-SW
File Tool Window Tools Window Help
Layout
Acquisition
Viewer
cellSens
LSM Imaging
Ocular
Scan Settings
Scanner:
Type: Galvano Resonant
Mode: OneWay Roundtrip
Interlace: OFF 2x 4x
Speed: 0.067 us/pixel
Image Size:
Aspect Ratio: 1:1 4:3
Scan Size: 512x512
High voltage correction: ON OFF
Pixel: 0.067 usec Line: 0.127 msec Frame: 65.779 msec
Area Settings
Rotation : deg Reset
Pan X : um Reset
Pan Y : um Reset
PMT Setting
PMT
Mode : VBF Lambda
Average: None Line Frame Times
Sequential Scan: None Line Frame
Dye & Detector Select
Phase1
Confocal Aperture Auto 120 um
Airy Disk x1.00
HSD1 HSD2
Group 1 CH1 CH2
Laser ND Filter: None 10%
P1 CH1 Violet HSD1 430 -470 nm G-1
405 0.3 %
HV 521 V
Gain 1.000 x
Offset 4 %
P1 CH2 Alexa Fluor 568 HSD2 570 -670 nm G-1
Live Graph Table Analysis Neuron Neuron_0001 Neuron_0002
Analysis ROI:
LUT: Hi-Lo Auto Contrast Reset
Tile Single
CH1 Violet CH2 Alexa Fluor...
CH1 Violet, CH2 Alexa Fluor 568
30 µm
Size:1024x1024 0.207[um/pixel] x,y=(0,142) Int:221, 238
Series
LSM
Time : ON OFF
Z : ON OFF
Series: 65.779 msec
Time Lapse
LSM Total: 0:00:00.07
Interval: FreeRun
Scan: 0:00:00.07 Rest: 0:00:00.00
Cycle: 10
Z Section
Motor: Start/End Range
Current:
Origin: 200.00
Register
Move
Slices: 12
Step Size: 0.83
Optimize
Start: 3703.46 Register
End: 3694.33 Register
Acquire
Normal Sync Sequence MATL
Imaging
LSM Start
SERIES DONE Append
APPEND NEXT 1
Bleach
Stimulation Start
LSM D:\CFC
Neuron
Current scan condition
Total scanning time : 0:00:00.07
Remaining time : -
Next scan start at : -
Neuron.oir
XY
點選"Resonant" 1 可將掃描器切換至高速模式
選擇"OneWay" 2 時，速度可達每秒鐘15張
此時影像尺寸最大值會固定在512x512 3
上午 10:51
2021/7/20


OLYMPUS FV31S-SW
File Tool Window Tools Window Help
Layout
Acquisition
Viewer
cellSens
LSM Imaging
Ocular
Scan Settings
Scanner:
Type: Galvano Resonant
Mode: OneWay Roundtrip
Interlace: OFF 2x 4x
1
Speed: 0.067 us/pixel
Image Size:
Aspect Ratio: 1:1 4:3
Scan Size: 512x512
High voltage correction: ON OFF
Pixel: 0.067 usec Line: 0.063 msec Frame: 33.333 msec
Area Settings
Rotation : deg Reset
Pan X : um Reset
Pan Y : um Reset
PMT Setting
PMT
Mode : VBF Lambda
Average: None Line Frame Times
Sequential Scan: None Line Frame
Dye & Detector Select
Phase1
Confocal Aperture Auto 120 um
Airy Disk x1.00
HSD1 HSD2
Group 1 CH1 CH2
Laser ND Filter: None 10%
P1 CH1 Violet HSD1 430 -470 nm G-1
405 0.3 %
HV 521 V
Gain 1.000 x
Offset 4 %
P1 CH2 Alexa Fluor 568 HSD2 570 -670 nm G-1
Live Graph Table Analysis Neuron Neuron_0001 Neuron_0002
Analysis ROI:
1:1 85 % LUT: Hi-Lo Auto Contrast Reset
Tile Single
CH1 Violet CH2 Alexa Fluor...
CH1 Violet, CH2 Alexa Fluor 568
30 µm
Size:1024x1024 0.207[um/pixel] x,y=(0,142) Int:221, 238
Series
LSM
Time : ON OFF
Z : ON OFF
Series: 33.333 msec
Time Lapse
LSM Total: 0:00:00.03
Interval: FreeRun
Scan: 0:00:00.03 Rest: 0:00:00.00
Cycle: 10
Z Section
Motor: Start/End Range
Current:
Origin: 200.00
Register
Move
Slices: 12
Step Size: 0.83
Optimize
Start: 3703.46 Register
End: 3694.33 Register
Acquire
Normal Sync Sequence MATL
Imaging
LSM Start
SERIES DONE Append
APPEND NEXT 1
Bleach
Stimulation Start
LSM D:\CFC
Neuron
Current scan condition
Total scanning time : 0:00:00.03
Remaining time : -
Next scan start at : -
Image List
07/20/2021 10:22:57.448 AM
XY
Neuron_0001.oir
選擇“RoundTrip” 1 時，速度可達每秒鐘30張
上午 10:51
2021/7/20


OLYMPUS FV31S-SW
File Tool Window Tools Window Help
Layout
Acquisition
Viewer
cellSens
LSM Imaging
Ocular
Scan Settings
1
Scanner:
Type: Galvano Resonant
Mode: OneWay Roundtrip
Interlace: OFF 2x 4x
Speed: 0.067 us/pixel
Image Size:
Aspect Ratio: 1:1 4:3
Scan Size: 512x512
High voltage correction: ON OFF
Pixel: 0.067 usec Line: 0.063 msec Frame: 9.759 msec
Area Settings
Rotation : deg Reset
Pan X : um Reset
Pan Y : um Reset
PMT Setting
PMT
Mode : VBF Lambda
Average: None Line Frame Times
Sequential Scan: None Line Frame
Dye & Detector Select
Phase1
Confocal Aperture Auto 120 um
Airy Disk x1.00
HSD1 HSD2
Group 1 CH1 CH2
Laser ND Filter: None 10%
P1 CH1 Violet HSD1 430 -470 nm G-1
405 0.4 %
HV 480 V
Gain 1.000 x
Offset 4 %
P1 CH2 Alexa Fluor 568 HSD2 570 -670 nm G-1
Series
LSM
Time : ON OFF
Z : ON OFF
Series: 9.759 msec
Time Lapse
LSM Total: 0:00:00.97
Interval: FreeRun
Scan: 0:00:00.01 Rest: 0:00:00.00
Cycle: 100
Z Section
Motor: Start/End Range
Current:
Origin: 200.00
Register
Move
Slices: 12
Step Size: 0.83
Optimize
Start: 3703.46 Register
End: 3694.33 Register
Acquire
Normal Sync Sequence MATL
Imaging
LSM Start
SERIES DONE Append
APPEND NEXT 1
Bleach
Stimulation Start
LSM D:\CFC
Neuron
Current scan condition
Total scanning time : 0:00:00.97
Remaining time : -
Next scan start at : -
Live Graph Table Analysis Neuron Neuron_0001 Neuron_0002 Neuron_0003
Analysis ROI:
Live Live x2 Live x4 1:1 76 % LUT: Hi-Lo Auto Contrast Reset
Tile Single
CH1 Violet
CH1 Violet
1R
2
CH2 Alexa Fluor 568
1R
CH1 Violet, CH2 Alexa Fluor 568
1R
Image List
07/20/2021 10:22:57.448 AM
Neuron_0001.oir
若想進一步提高掃描速度，可以點選此處 1 ，在畫面
上畫出掃描區域 2 ，即可獲得更高的fps，將尺寸縮小
至512x32時，可達極速438 fps
上午 10:57
2021/7/20

OLYMPUS FV31S-SW

File Tool Window Tools Window Help

Layout | Acquisition | Viewer | cellSens

LSM Imaging | Ocular

Scan Settings

Delete All ROIs for Imaging

Scanner:
Type: Galvano Resonant
Mode: OneWay Roundtrip
Interlace: OFF 2x 4x
Speed: 0.067 us/pixel
Image Size:
Aspect Ratio: 1:1 4:3
Scan Size: 512x512
High voltage correction: ON OFF
Pixel: 0.067 usec Line: 0.063 msec Frame: 9.759 msec

Area Settings

Rotation : 0.0 deg Reset
Pan X : 0.00 um Reset
Pan Y : 0.00 um Reset

PMT Setting

PMT

Mode : VBF Lambda
Average: None Line Frame 2 Times
Sequential Scan: None Line Frame
Dye & Detector Select
Phase1
Confocal Aperture Auto 120 um
Airy Disk x1.00
HSD1 HSD2
Group 1 CH1 CH2
Laser ND Filter: None 10%
P1 CH1 Violet HSD1 430 -470 nm G-1
405 0.4 %
HV 480 V
Gain 1.000 x
Offset 4 %
P1 CH2 Alexa Fluor 568 HSD2 570 -670 nm G-1

Live | Graph | Table | Analysis | Neuron | Neuron_0001 | Neuron_0002 | Neuron_0003

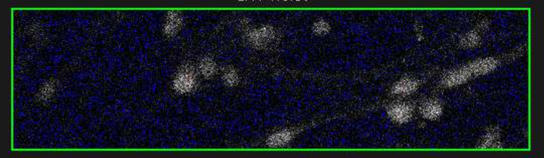
Analysis ROI:

Live | Live x2 | Live x4 | 1:1 100 % | LUT: Hi-Lo | Auto Contrast | Reset

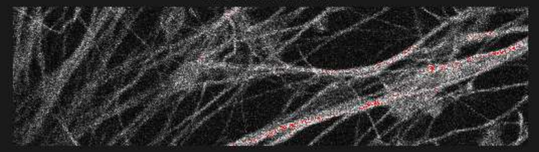
Tile | Single

CH1 Violet

CH1 Violet

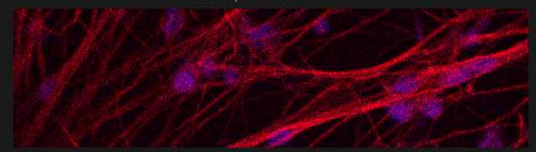


CH2 Alexa Fluor 568



如圖中所示，掃描此影像範圍可達100 fps
拍攝300張連續圖像只需要3秒鐘時間

CH1 Violet, CH2 Alexa Fluor 568



Size:512x138 0.414[um/pixel]

Series

LSM
Time : ON OFF
Z : ON OFF
Series: 9.759 msec

Time Lapse
LSM Total: 0:00:02.93
Interval: FreeRun
Scan: 0:00:00.01 Rest: 0:00:00.00
Cycle: 300

Z Section
Motor: Start/End Range
Current: 3707.72 Origin: 200.00
Register
Move
Slices: 12
Step Size: 0.83
Optimize
Start: 3703.46 Register
End: 3694.33 Register

Acquire

Normal | Sync | Sequence | MATL

Imaging
LSM Start
SERIES DONE Append
APPEND NEXT 1

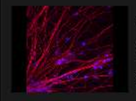
Bleach
Stimulation Start

LSM D:\CFC
Neuron

Current scan condition

Total scanning time : 0:00:02.93
Remaining time : -
Next scan start at : -

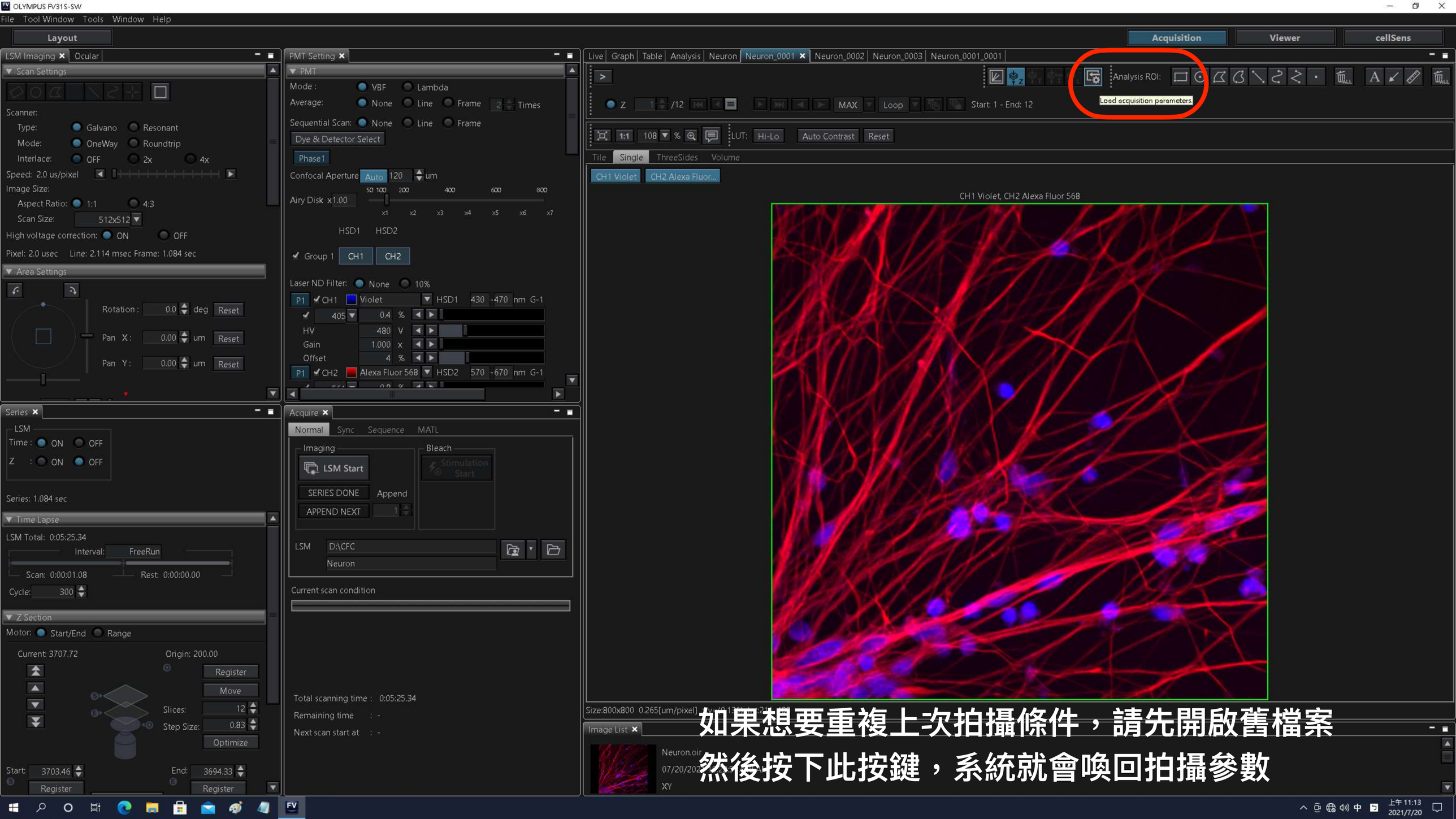
Image List



Neuron.oir
07/20/2021 10:22:57.448 AM
XY

Neuron_0001.oir

上午 10:58
2021/7/20


OLYMPUS FV31S-SW
File Tool Window Tools Window Help
Layout
Acquisition
Viewer
cellSens
LSM Imaging
Ocular
Scan Settings
Scanner:
Type: Galvano Resonant
Mode: OneWay Roundtrip
Interlace: OFF 2x 4x
Speed: 2.0 us/pixel
Image Size:
Aspect Ratio: 1:1 4:3
Scan Size: 512x512
High voltage correction: ON OFF
Pixel: 2.0 usec Line: 2.114 msec Frame: 1.084 sec
Area Settings
Rotation: 0.0 deg Reset
Pan X: 0.00 um Reset
Pan Y: 0.00 um Reset
PMT Setting
PMT
Mode: VBF Lambda
Average: None Line Frame 2 Times
Sequential Scan: None Line Frame
Dye & Detector Select
Phase1
Confocal Aperture Auto 120 um
Airy Disk x1.00
HSD1 HSD2
Group 1 CH1 CH2
Laser ND Filter: None 10%
P1 CH1 Violet HSD1 430 -470 nm G-1
405 0.4 %
HV 480 V
Gain 1.000 x
Offset 4 %
P1 CH2 Alexa Fluor 568 HSD2 570 -670 nm G-1
Series
LSM
Time: ON OFF
Z: ON OFF
Series: 1.084 sec
Time Lapse
LSM Total: 0:05:25.34
Interval: FreeRun
Scan: 0:00:01.08 Rest: 0:00:00.00
Cycle: 300
Z Section
Motor: Start/End Range
Current: 3707.72
Origin: 200.00
Register
Move
Slices: 12
Step Size: 0.83
Optimize
Start: 3703.46 Register
End: 3694.33 Register
Acquire
Normal Sync Sequence MATL
Imaging
LSM Start
SERIES DONE Append
APPEND NEXT 1
Bleach
Stimulation Start
LSM D:\CFC
Neuron
Current scan condition
Total scanning time : 0:05:25.34
Remaining time : -
Next scan start at : -
Live Graph Table Analysis Neuron Neuron_0001 Neuron_0002 Neuron_0003 Neuron_0001_0001
Analysis ROI:
Load acquisition parameters
Z /12 MAX Loop Start: 1 - End: 12
1:1 108 % LUT: Hi-Lo Auto Contrast Reset
Tile Single ThreeSides Volume
CH1 Violet CH2 Alexa Fluor...
CH1 Violet, CH2 Alexa Fluor 568
Size:800x800 0.265[um/pixel]
Image List
Neuron.oir
07/20/202
XY
如果想要重複上次拍攝條件，請先開啟舊檔案
然後按下此按鍵，系統就會喚回拍攝參數
上午 11:13
2021/7/20