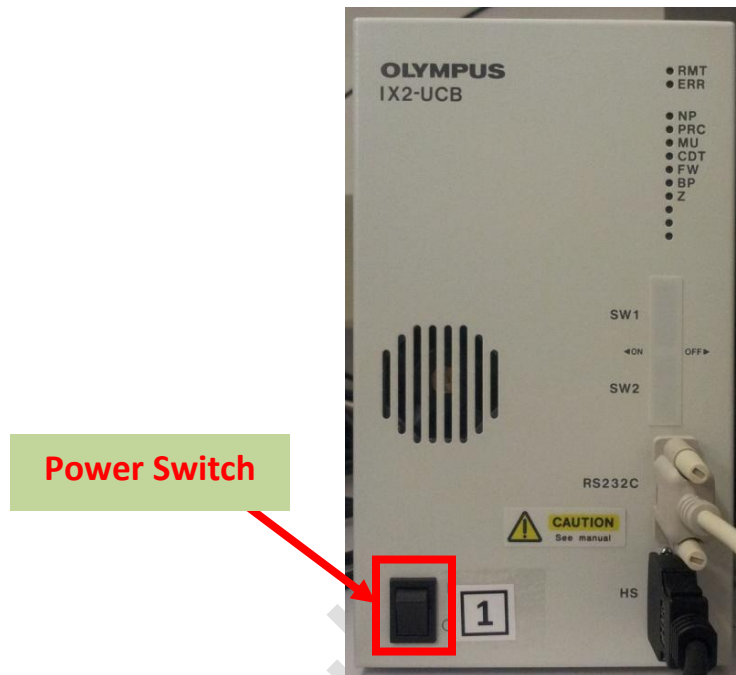


# Olympus Time-lapse Microscope

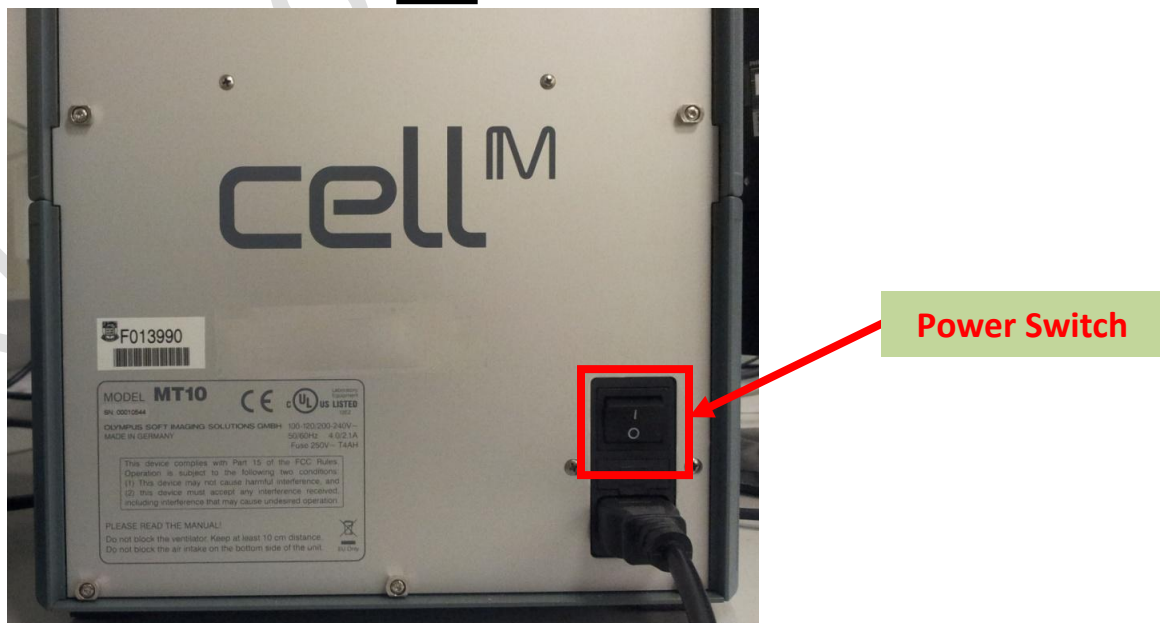
## Basic operation

### To start up the microscope

1. Switch on the Olympus UCB. (label as **1** )



2. Switch on the MT10. (label as **2** )



3. Switch on the camera. (label as **3**)



Power Switch

4. Turn on the Computer. (label as **4**)



5. Start “Xcellence Pro” in the computer.

(Continue on next page)

6. After started the “Xcellence Pro”, you will see the control panel.

The screenshot displays the Xcellence Pro software interface. On the left, a vertical list of image buffers (Image 1 to Image 17) is shown, with a red arrow pointing to the first buffer. The central area is a large viewport for viewing images or live camera feed. To the right, there are several control panels: a 'Camera control' panel with settings for exposure time and brightness, a 'Microscope' panel with controls for Z-drive, objectives, and filters, and an 'Illumination System MT' panel with controls for excitation, shutters, and intensity. Red arrows point from numbered callouts to these panels: 1 points to the image buffer list, 2 points to the central viewport, 3 points to the camera control panel, 4 points to the microscope control panel, and 5 points to the illumination system control panel.

1. Image Buffer

2. Viewport for viewing image or live view from camera

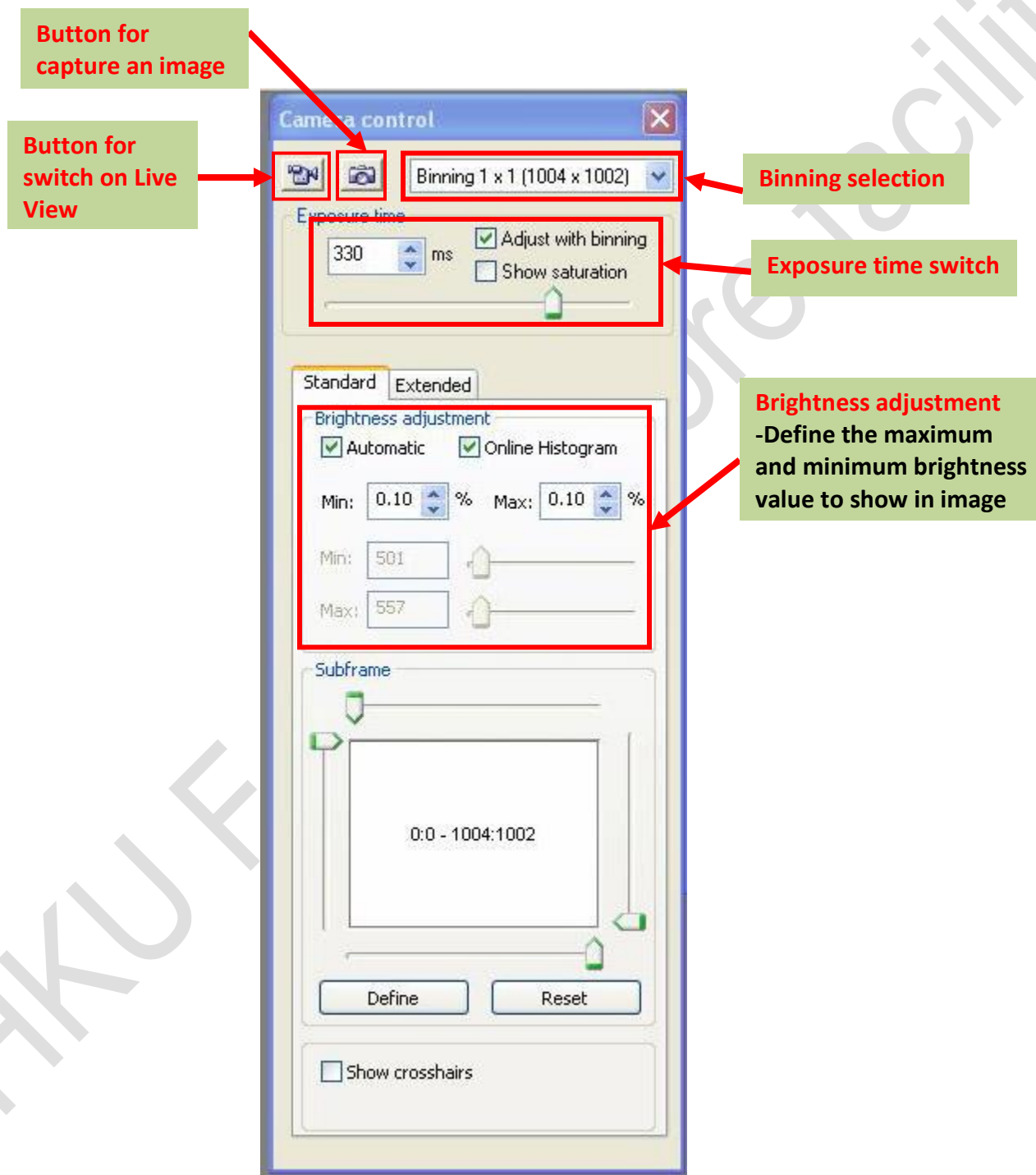
3. Camera control

4. Microscope control

5. illumination system control

## Introduction of the Xcellence Pro control panel

In “**3. Camera control**” panel, you can control the camera and capture image.



In “4. Microscope control” panel, you can control the microscope.

The screenshot shows the 'Microscope' control window. At the top, the 'Z-Drive' section displays a position of 1269.1  $\mu\text{m}$  with up/down arrows and a 'Change limits' button. Below this, the 'Objectives' section has a dropdown menu currently set to 'UPLFLN 4x'. The 'Fluorescence turret' section shows 'MGFP' selected. The 'Condenser' section has 'BF' selected. The 'Lamp' section features a slider set to 3.8. Below the lamp, there are two icons: a microscope eyepiece and a camera. At the bottom, there is a grid of filter set buttons: DIC, DAPI, GFP, Cy3, Cy5, BF, pos7, and pos8. Red arrows point from text boxes to specific controls: 'Objectives Switch' points to the objective dropdown; 'Condenser Switch' points to the condenser dropdown; 'Lamp Switch' points to the lamp slider; 'Lamp shutter Switch' points to the eyepiece/camera toggle icons; 'Imaging filter Switch' points to the filter set buttons; and 'Eyepiece & Camera Switch' points to the eyepiece/camera toggle icons.

**Objectives Switch**  
- Switch different objective

**Condenser Switch**  
- Switch different condenser

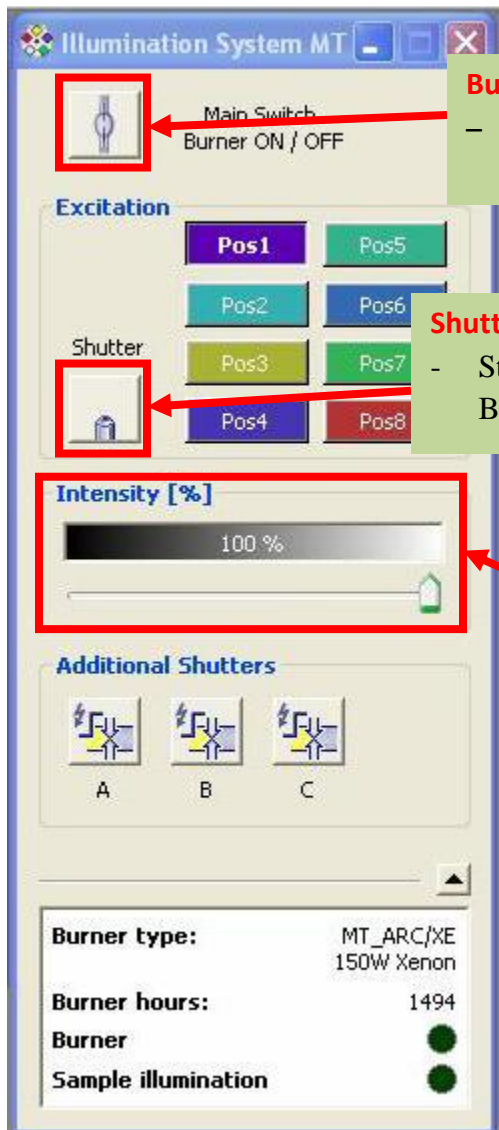
**Lamp Switch**  
- Switch to turn On/Off lamp and changing intensity

**Lamp shutter Switch**  
- Turn On/Off shutter to stop illumination

**Imaging filter Switch**  
- For user switch to different filter set

**Eyepiece & Camera Switch**  
- Toggle between the microscope eyepiece and camera

In “5. Illumination system control” panel.



The screenshot shows the 'Illumination System MT' control window. It features a 'Main Switch' icon (a light bulb) at the top left, a 'Shutter' icon (a camera shutter) below it, and an 'Intensity [%]' slider below that. The slider is currently set to 100%. Below the slider are three 'Additional Shutters' labeled A, B, and C, each with a circuit diagram. At the bottom, there is a status section with 'Burner type: MT\_ARC/XE 150W Xenon', 'Burner hours: 1494', and two green indicator lights for 'Burner' and 'Sample illumination'.

**Burner Switch**

- Start the “Burner” if you need to capture fluorescence image

**Shutter Switch**

- Stop the excitation for a short period to avoid Bleaching of your sample

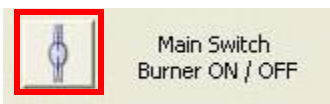
**Intensity Switch**

- Adjust the burner intensity level



## To capture fluorescence multi-channel image

1. Switch on the “**burner**” in “**5. Illumination system control**” panel if you need to capture fluorescence image.



2. Switch on the “**lamp**” in “**4. Microscope control**” if you need to capture DIC image.



3. Switch the microscope view to “**camera**” in “**4. Microscope control**”.
4. Select “**live view**” in “**3. Camera control**”.

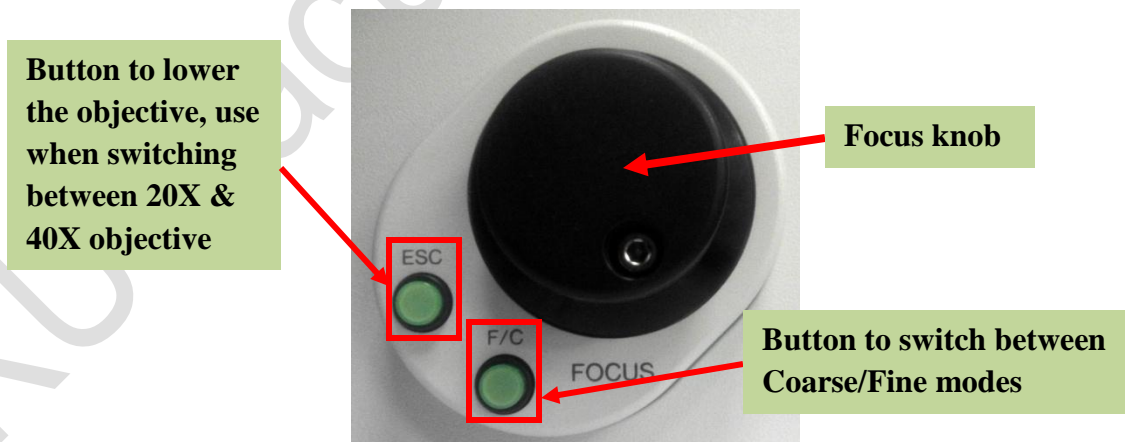


5. Select the “**image filter**” you need to use for your image in “**4. Microscope control**”.



6. Select the objective you want and get the correct focus of your image.

To focus your image, use the **focus knob** at the microscope stand.



7. Click “**Capture**” to get your image, the captured image will store at “**image buffer**”.



8. Repeat Step 4 to Step 7 to capture another image.
9. To merge the images you have captured, select one of the images you want to merge in “**image buffer**”, then select “**Image → combine**”. Select the image(s) you need to merge and then click OK. The merged image will be stored at “**image buffer**”.



10. To save your image(s) store at image buffer, select the image want to save and then choose “**File → Save as**” to save your image.

**(Please save your file at “D:/User/your name”)**

### **To use experiment manager to capture fluorescence multi-channel, time lapse and Z-stack image**

1. Open your **database** by select “**Database → Open**”.  
(If you do not have a database, select “**Database → Administration → New database**”.)  
In the New database window, enter your name and save your database in “**D:/User/your name**”, Then press “**Next**” followed by “**Finish**”)

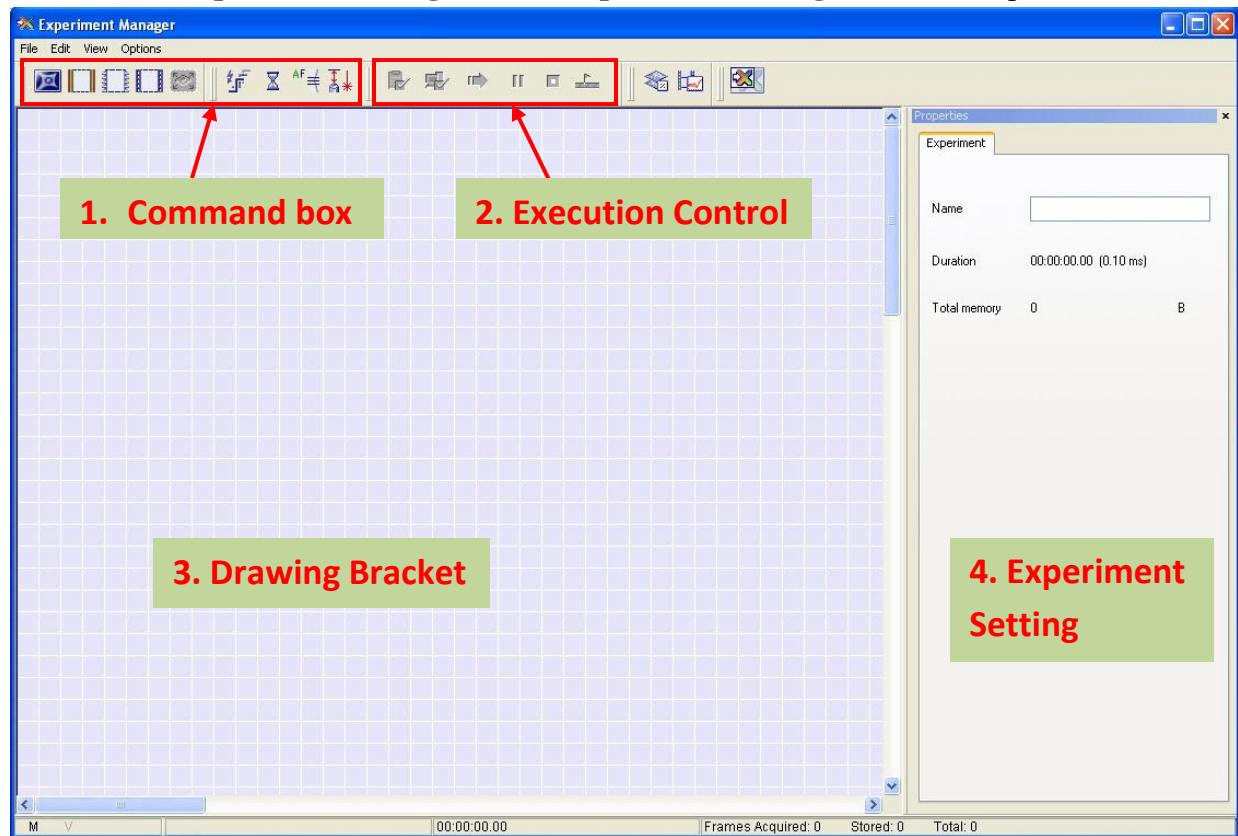


2. Open the “**Experiment Manager**”.





3. After select “**Experiment Manager**”, the “**Experiment Manager Panel**” is open.



### 1. Command Box





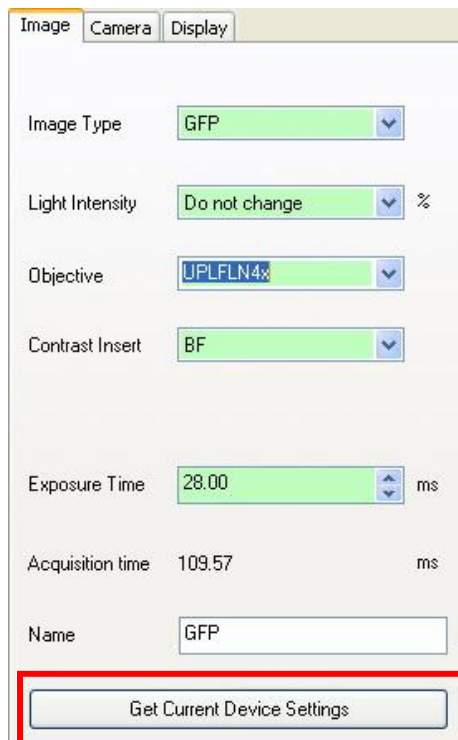
### 2. Execution Control




4. To design the image experiment, drag the “**command box**” into the “**3. Drawing bracket**”.

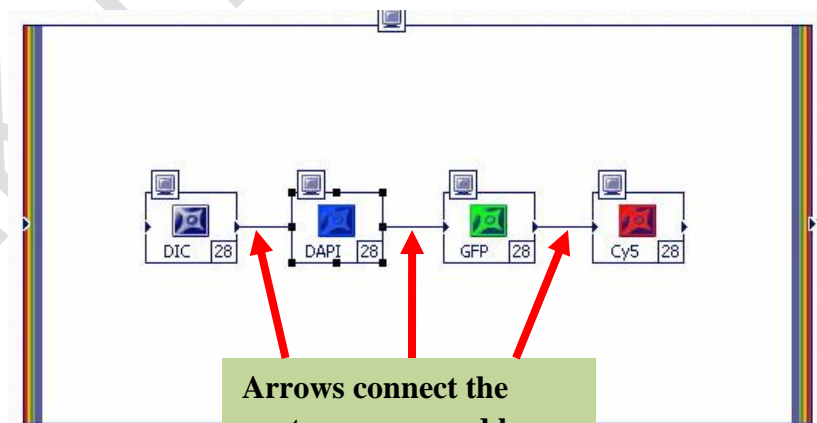
**4a/ For multi-color imaging**

- i. Click and drag  into drawing bracket. E.g. If you need to have 4 color images, drag 4  into the drawing bracket. Connect the command with arrow(s).
- ii. Set the image capture value in the experiment setting for each command.



You can use “Get Current Device setting” to get the current capture value

- iii. Click and drag the  multi-color frame into the drawing bracket, the frame should include all the capture command boxes.





Multi-color frame should include all the command boxes

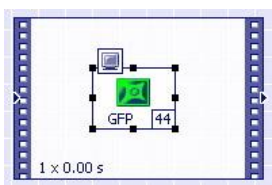
Arrows connect the capture command boxes

#### **4b/ For Time-lapse imaging**

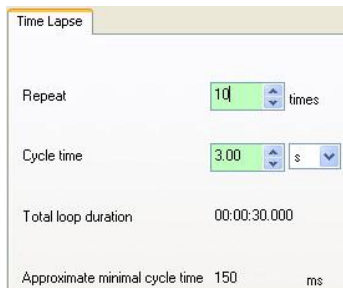
(For time-lapse imaging, you may need to use the incubator, the steps to install the incubator at page 14)



- i. Click and drag  into drawing bracket. Change the setting at the “**4.experiment setting**”.

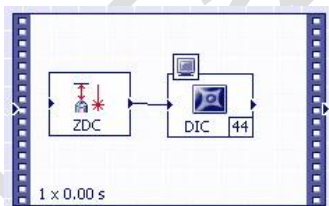
- ii. Click and drag the  **time lapse frame** into the drawing bracket, the frame should include all the capture command boxes.



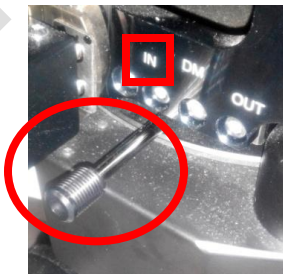
- iii. Click the time lapse frame; change the setting in the “**4.experiment setting**”.





- iv. User can also use  **auto-focus** in time-lapse capture. Click and drag  into the drawing bracket. Connect the command with arrow(s).

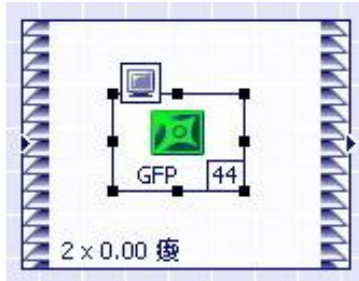


(The auto-focus only available for **20X and 40X objective** only. Also ensure you have pushed the ZDC stick at the microscope stand to “**in**” position.)

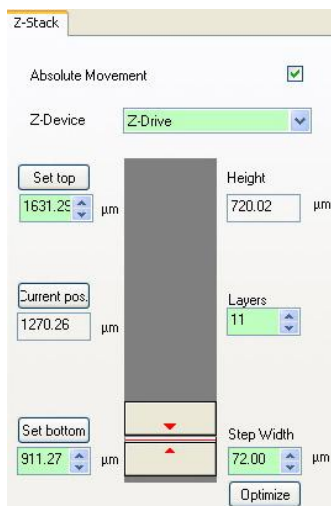


#### **4c/ For Z-stack imaging**

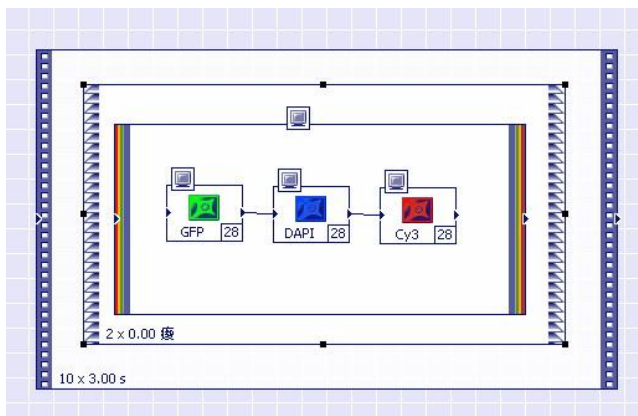
- i. Click and drag  into drawing bracket. Change the setting the experiment setting.
- ii. Click and drag the  **Z-stack frame** into the drawing bracket, the frame should include all the capture command boxes.





- iii. Click the **Z-stack frame**; Change the setting in the “**4.experiment setting**”.





User can also make different combination of experiment command they need.



5. After drawing the experiment plan, select  to **verify** the experiment plan. If the plan has error, message will be display. Please modify the plan and verify again.

6. If the plan does not have any problem, select  to allow the system to **prepare** the experiment.

7. Select  to **run** the experiment plan after preparation is complete.

When the experiment plan is running, you can “ **Pause**” or “ **Stop**” the plan any time.

8. After the experiment plan finish, you image file will be store at the “1.image buffer”.  
To save your image at image buffer, select the image want to save and then choose “**File**→**Save as**” to save your image.

For time-lapse video, you can save the video by choosing “**File** → **Export to AVI**”.

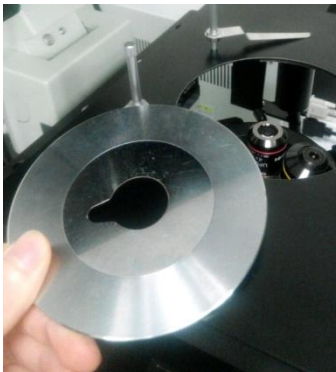
**(Please save your file at “D:/User/your name”)**

## **To install the incubator for time-lapse imaging**

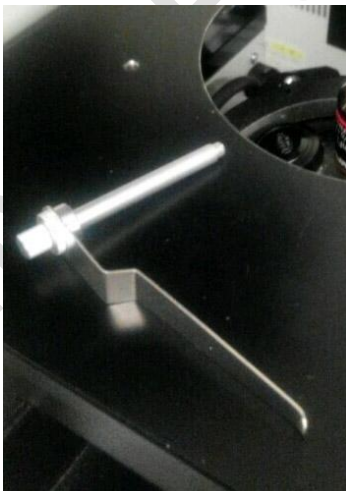
1. Raise the microscope arm **gently**.



2. Remove the metal plate from the stage.



3. Remove the slide clips from the stage.





4. Get the **incubator** from the plastic tray (**Label as A**), put it on the stand. It should fit into the hole on the stage.



5. Use the key to install the screws in order to fix the incubator on the stage.

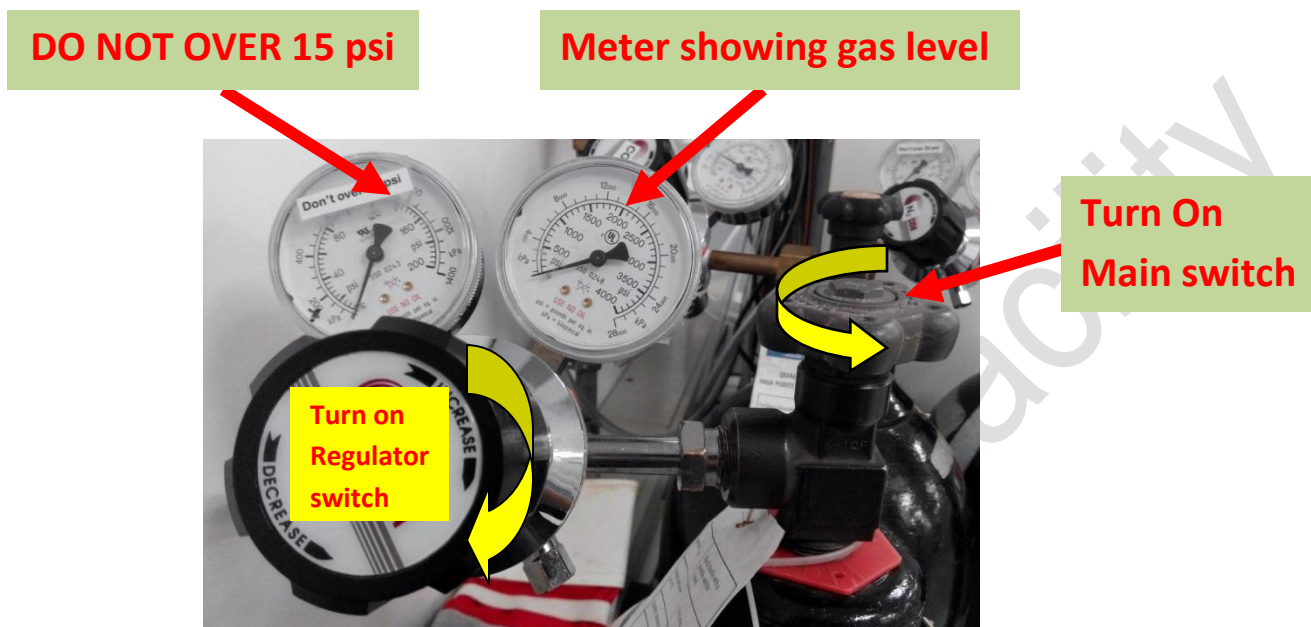


6. Add **50mL autoclaved water** into the incubator with the falcon tube **carefully**.
7. **Switch on the heater. (Label as B) Do not change any setting of the heater.**



Power Switch

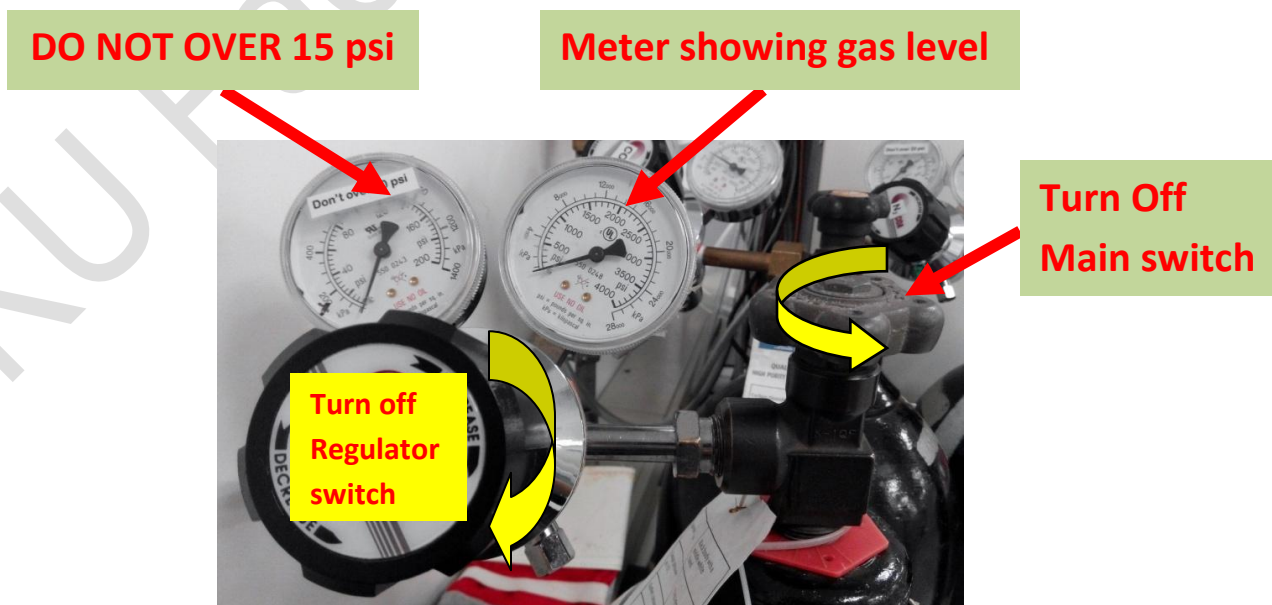
- Turn on the CO<sub>2</sub> mixture gas under the table. **The gas flow pressure should set at 15 psi.**



- Place your culture dish into the incubator. **30 minutes is needed** to allow the temperature reaching equilibrium.
- Lower the microscope arm gently.

### To remove the incubator after time-lapse imaging

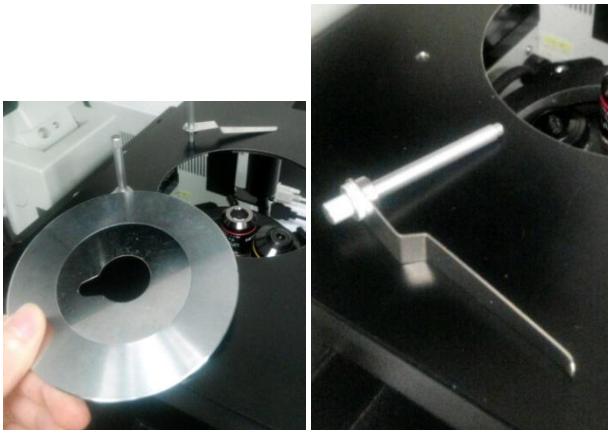
- Switch off the heater. (Label as B) Do not change any setting of the heater.**
- Turn off the CO<sub>2</sub> mixture gas under the table.**



3. Use the key to remove the screws in order to un-mount the incubator on the stage.



4. **Remove the water from the incubator carefully.** Use the plastic container to collect the water. Use paper towel to dry the incubator.
5. Put the incubator back to the tray (**Label as A**).
6. Install the **metal plate and slide clips** on to the stage.



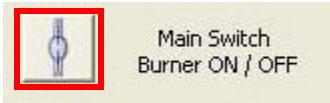
7. Lower the microscope arm gently.

## **To turning off the microscope**

1. Ensure you have saved all the images in the image buffer.

**All images in the buffer will be deleted after the Xcellence program is closed.**

2. **Turn off the burner before closing the Xcellence program. (Important!)**



3. Close the Xcellence program.
4. Shut down the computer.
5. Switch off the **Olympus UCB**. (label as **1**)
6. Switch off the **MT10**. (label as **2**)
7. Switch off the **camera**. (label as **3**)
8. **Sign the log sheet** before you leave.

**End**