1. **Select a vessel.**
   In the VESSEL panel, click **Select** to bring up the vessel selection window and select a vessel.

2. **Select an objective.**
   In the CHANNELS panel, click the desired objective to select the corresponding magnification.
   **IMPORTANT!** If using an objective with a correction collar, adjust the correction collar as necessary according to the bottom thickness of the selected vessel.

3. **Set up the channels.**
   In the CHANNELS panel, set up channels as needed.
   **CAUTION!** The CELENA® X uses Class 3B ultraviolet LEDs that are in accordance with IEC/EN 60825-1. Make the door is closed when imaging.
   **IMPORTANT!** Minimize the time that the sample is being exposed to light to prevent photobleaching and/or phototoxicity.

   **Select a channel.** Click the desired channel and adjust the condenser’s iris diaphragm.
   - For brightfield imaging, click **BF/PH** for transmitted light and adjust the iris diaphragm as desired.
   - For fluorescence imaging, click the desired fluorescence channel.

   **Adjust light intensity.** Move the slider in the desired direction or enter the desired value in the text box.

   **Adjust camera gain and exposure.** Move the slider in the desired direction or enter the desired value in the text box.
   **QUICK TIP** When searching for a sample, increase gain and decrease exposure for a faster frame rate. For imaging, decrease gain and increase exposure to reduce background noise.

   **Focus sharply.** Move the focus slider in the desired direction or enter the desired value in the Z-position box. Alternatively, click **Find Focus** to have the CELENA® X find the optimal focal plane.
   **NOTE** The speed of the image-based autofocus is entirely dependent on the set exposure. Reducing the exposure to < 10 ms will increase focusing speed.

   **Set up Multiscan AF.** Make sure the current field is focused sharply before proceeding. Click **Multiscan AF.** Select how often to autofocus during an automated scan. Select whether to use the image-based or laser autofocus.
   - **Image-based:** Set the range to scan from the current focal position. Turn the user-defined focus offset on or off.
   - **Laser AF:** Click **Pre-Scan** to configure the laser autofocus settings. Click **Test AF** to test the accuracy of the autofocus. Laser AF cannot be used with magnifications below 10X.
   **NOTE** When using this feature for imaging in multiple channels, Multiscan AF must be set for each channel. This is especially important when the fluorescent markers in different channels are in different focal planes.

   **Add to the project protocol.** Click **Add.**
   Repeat for all necessary channels.
This Quick Start Guide provides instructions for well plate imaging with the CELENA® X High Content Imaging System. For detailed instructions, refer to the user manual in the supplied USB drive.

4. Select areas and fields to be captured.
In the VESSEL panel, select the well(s) to image in the vessel map by clicking individual wells or dragging and dropping to select multiple wells. Select the field(s) to image in the well map by clicking individual fields or dragging and dropping to select multiple fields.

5. (Optional) Set up Z-stack imaging.
In the Z-STACK panel, select a Z-stack method and set appropriately.
- **Start/End**: Move the focal plane to the desired start Z-position and click Set Start. Move the focal plane to the desired end Z-position and click Set End. Select to capture images at specific intervals (µm) or to capture a specific amount of images (steps) and enter the desired value.
- **Range (current)**: Move the to the desired start position. To set the imaging range, enter how far above (+) and below (-) the current position to set the imaging range. Select to capture images at specific intervals (µm) or to capture a specific amount of images (steps) and enter the desired value.
- **Range (AF)**: Make sure Multiscan AF has been set. To set the imaging range, enter how far above (+) and below (-) the autofocused position. Select to capture images at specific intervals (µm) or to capture a specific amount of images (steps) and enter the desired value.

Click Add.

6. (Optional) Set up time lapse imaging.
In the TIME LAPSE panel, set the total imaging time and interval.

- **IMPORTANT!** Set up time lapse imaging last so that the CELENA® X can account for the other imaging options you have set, which affect the time required to capture one image.

Click Add.

7. Create a project.
In the PROJECT panel, click Create Project. Name the project and designate where to save the project folder.

- **IMPORTANT!** Save projects on the computer from which you are running Explorer. Do not save the project to an external hard drive or a USB drive as this can affect imaging time.

8. Run the project.
Once a protocol has been set and project has been created, click RUN at the bottom of the PROJECT panel.

- **IMPORTANT!** Make sure the CELENA® X door is closed for fluorescence imaging applications to block ambient light and improve fluorescence image quality.

To pause a running project, click PAUSE.
To stop a running project, click STOP.