

# ProtoCOL

Colony Counting and Zone Sizing System

## Quick Start Guide - Inhibition Zones

### Starting ProtoCOL

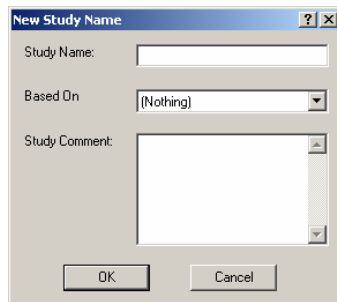
1. Connect ProtoCOL to a monitor keyboard and mouse, and switch on via the button at the back of the unit. This will start the internal PC and initialise the internal camera.
2. Double click the ProtoCOL icon on the desktop. If required enter your specific user name and password, otherwise ProtoCOL will start automatically.
3. When ProtoCOL opens for the first time, the Applications Window will open but will be empty. On subsequent occasions the same will happen but the window will open showing the same information as the last time it was closed.

### Data storage in ProtoCOL

Data is stored in **STUDIES** and **BATCHES** with each study being divided into a series of batches. Each batch must contain results from the same type of plate although a study may contain different batches with different types of results. Before taking measurements using ProtoCOL you will need to have a study file open. You can either open an existing file or create a new one. ProtoCOL will automatically save results to an open study file.

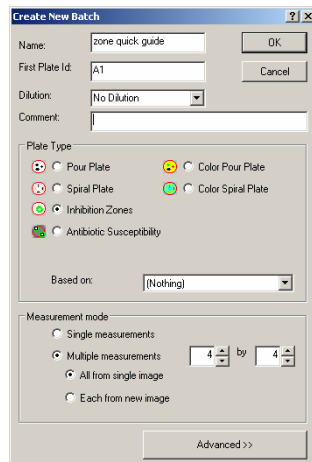
### Creating a new study and a new batch

1. Choose NEW from the File menu, or click the "New Study" button in the standard tool bar.
2. Enter a name for the new study.



3. If you are going to use existing batch definitions for your new study, choose the name of the file from the "Based On" drop down menu. Or choose "Nothing" for a brand new study.
4. If required enter a comment in the Study Comment box.
5. Click OK to confirm the settings and close the dialog box.

If you choose to base your new study on an existing one the process of creating a new study is complete. However, if you are creating a completely new study you now need to create at least one new batch to go in it. The "Create New Batch" dialog box will open automatically if you do not base your new study on an existing one. To create a new batch within an existing study choose "New Batch" from the Edit menu.

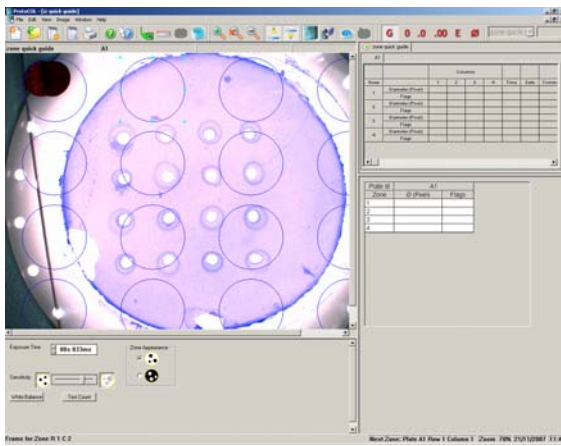


Starting ProtoCOL

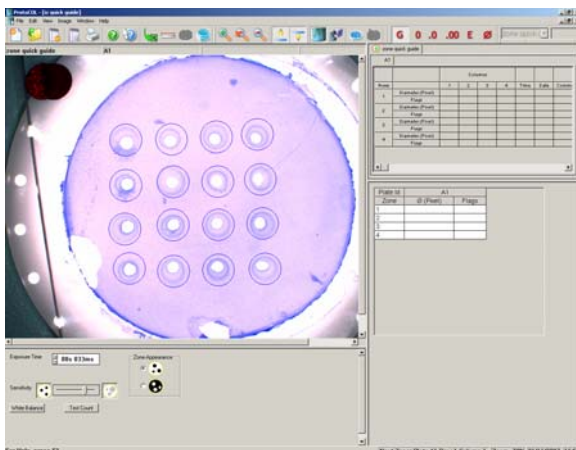
Data storage in ProtoCOL

Creating a new study and a new batch

6. Enter a name for the new batch.
7. Enter an ID for the first plate in the batch.
8. Choose inhibition zones from the list of measurement types
9. If required enter a comment for the batch.
10. If required choose a set of previously saved settings from the appropriate "Based On" drop down menu.  
If you do this the new batch is now created. If this is a brand new batch click "Nothing" from the drop down menu.
11. If this is a completely new batch choose "multiple images" and then all from "single image". From here select the number of rows and columns by using the up and down arrows.  
To set more advanced options click the Advanced Option button to expand the dialog box.
12. In the "Saved Image Options" box check the Automatic option to save the displayed image automatically after taking a measurement.
13. When all of the information is complete click "Hide" to shrink the dialog box and OK to close it and accept the new settings.  
The new batch is now created and the following screen will be displayed.

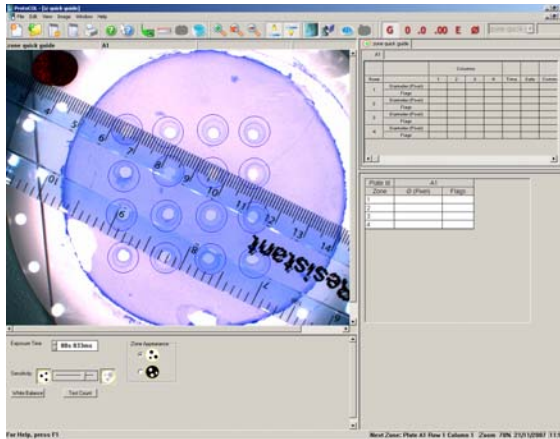


14. Use the zoom buttons so that the image appears correctly on the screen. To re-size the measurement frames, click the left mouse button within one of the frames so that the "Drag Handles" appear. Position the cursor over one of the drag handles and while holding the left mouse button down move the cursor so that the counting zones are re-sized. Re-position the zones by holding the left mouse button down within the frame that you want to move and re-positioning it to the required zone.



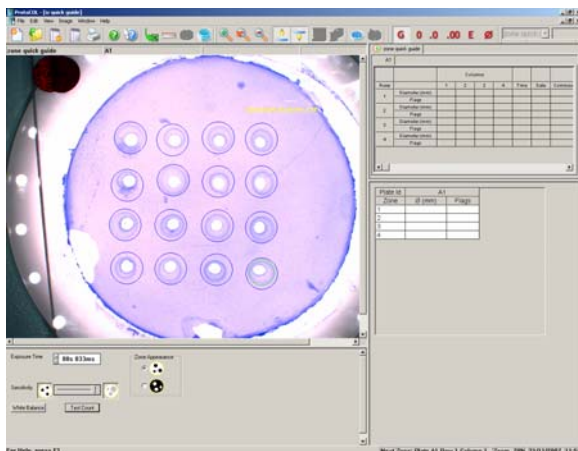
### Calibrating ProtoCOL and setting up batches to take measurements

1. Choose the light setting by using the up-light and down-light buttons in the image tool bar. For clear agars with good contrast colonies best results are usually obtained using the lower lamp. Before using ProtoCOL to count plates you will need to perform an individual calibration. If you try to use an uncalibrated study you will be reminded that calibration is required.
2. To calibrate ProtoCOL insert an object of known size or with known graduations on it within the camera's field of view.
3. Choose "Calibration" from the edit menu or use the calibration button on the tool bar.
4. Move the pointer to the starting point on the object and click the left mouse button. As you move the pointer along the object a line will be drawn behind it to the final position. When you reach the end at which you decide to take the calibration measurement click the left mouse button again.
5. The calibration dialog box will be displayed. Enter the distance between the two points and the units used for the measurement.
6. Press OK to confirm the calibration and close the dialog box.

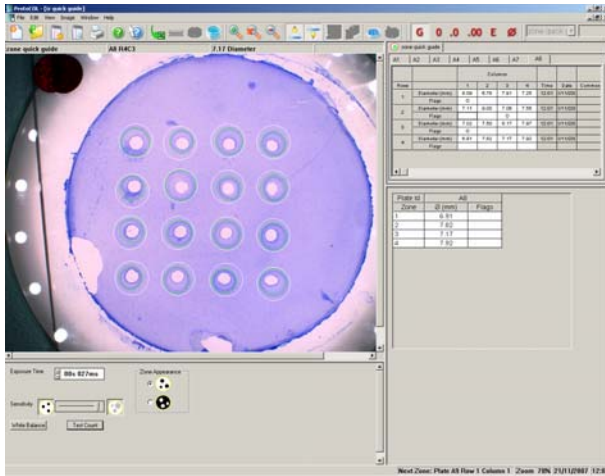


### Optimising camera settings

1. Place a typical plate for the batch under the camera. ProtoCOL will automatically capture an image of the plate and display it in the image pane.
2. Adjust the camera gain / sensitivity and exposure time using the slider and spin arrows as appropriate. The camera gain slider should be set to the left to give a low value.
3. Set the colour correction of the camera by placing a piece of white paper under the camera and clicking on "White Balance". This is now complete and should not have to be done very often.
4. Use the controls in the "Controls Pane" to select the appearance of the plate, either light zones on a dark agar or dark zones on a light agar.
5. Adjust the sensitivity by moving the slider. As these changes are made you can see in real time the difference that this will have on the measurement made by ProtoCOL.
6. Click "Test Count".
7. The "Test Zone" that has been read is highlighted in green. Carefully adjust the camera settings again to achieve the most accurate result.



8. Click the "Count Colonies" icon.
9. Results are shown in the top right of the screen along with information on time and date that the plate was read, dilution factor, batch name and any observation. The study name is displayed on the top left of the screen.



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