

# aCOLyte3

## User Manual



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# Getting started

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## Introduction

aCOLyte3 is a combined hardware and software system for automatically counting colonies on plates.

Once the system has been set up, all you have to do to count the colonies is to put each plate on the plate holder and press a button. The counting of each plate takes no more than a few seconds and the results are shown instantly in a table.

aCOLyte3 allows you to enter plate identifiers and dilutions manually, to use auto-incrementing plate identifiers or to enter lists of plate identifiers and dilutions within aCOLyte3 or to import them from a CSV file. If you use auto-incrementing plate identifiers or a plate list, aCOLyte3 will prompt you for which plate to insert next. The plate identifiers and dilutions are then automatically assigned to the results and recorded. You can also read plate identifiers and dilutions using a barcode reader.

When you are counting colonies, you can use aCOLyte3:

- to apply sophisticated algorithms to distinguish touching colonies;
- to add or remove colonies manually after an automatic count – you can also perform a completely manual count if required;
- to define ‘exclude regions’ on the plate if there are any problem areas on an individual plate where colonies cannot be distinguished for some reason – the missing area is then automatically allowed for in calculating the total count for the plate.

All counting results are displayed and saved automatically, and any manual changes you make to a result are marked against the individual result with comments and coded flags. By default, an image of the plate is automatically saved with each result, and can be reloaded at a later time for auditing. You can also view a full audit history for any changes you have made to a result.

Once you have completed the counts/measurements, you can compile a report showing the results.

This introduction has only mentioned a few of the features that make aCOLyte3 such a powerful but easy tool to use. The rest of the Manual gives full instructions for using all of its many functions.

## Structure of the Manual

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**Note** The cross-references in this manual assume that it is printed or viewed with odd numbered pages on the right-hand side. If you are using Adobe Acrobat to read this manual on-screen, you can display two pages side-by-side by choosing **Two Page View** from the **View→Page Display** submenu, and you can then choose **Show Cover Page in Two Page View** from the same submenu to display the odd numbered pages on the right-hand side.

---

The rest of this Manual is structured as follows:

- The next section, *Using aCOLyte3 – overview* in this *Getting Started* chapter provides a brief overview of the way you use aCOLyte3 to count colonies. This involves two main phases:
  - using *the Batch Designer* to set up the system for detecting and counting colonies on a series of plates;
  - using *Measurement mode* to actually count the colonies on the plates and record the results.

The next section also introduces you to the idea of a ‘batch’, which is used to hold the settings made in *the Batch Designer* and then to store the results produced in *Measurement mode*.

The final two sections of this *Getting Started* chapter give instructions for starting aCOLyte3 (see *Starting aCOLyte3*, on the facing page) and then for logging out and closing aCOLyte3 (see *Logging out and closing aCOLyte3*, page 5).

The remaining chapters are:

- *Capturing images*, page 7, which shows you how to load plates into aCOLyte3 and capture an image of the plate.

These procedures are used in the Batch Designer and Measurement mode.

- *Creating a new batch*, page 17, which shows you how to use the Batch Designer to create a new batch and specify how colonies should be counted.

Some of these procedures can also be applied in Measurement mode.

- *Using Measurement Mode to count colonies*, page 61, which shows you how to count the colonies on a series of plates.
- *Working with results*, page 77, which shows you how to view and edit the results of your measurements and how to create reports using the results.
- *Configuring aCOLyte3*, page 91, which shows you how to configure aCOLyte3 in a variety of ways, including how to set up and manage user accounts to protect access to the program.

## Using aCOLyte3 – overview

aCOLyte3 uses ‘batches’ to define the settings used to detect colonies on plates and to store the results of the measurements you make using those settings. This means that before you can use aCOLyte3 to take any measurements, you must have an open batch. You can do this by opening an existing batch or by creating a new batch. You can create a completely new batch or base a new batch on an existing one, either to use the same settings, or as a starting point for defining new settings. aCOLyte3 automatically saves your results in the current batch, so once you have created and accepted a batch design, you do not need to take any action to save it again.

Each batch contains the settings and results for detecting and counting the colonies on a series of plates that have the same types of colony, use the same medium and are of the same type (Pour Plate or Spiral Plate).



### Creating a batch

In many cases, all you need to do to set up the colony detection procedure for a batch is specify whether the colonies are lighter or darker than the background, but if required, you can also fine tune the colony detection by adjusting the sensitivity, setting a minimum size for colonies and choosing whether to use the colony splitting algorithm.

When you create the batch, you also need to choose how plate identifiers and dilutions are entered for each plate as you measure it. If required, you can also enter count restrictions so that warnings are given if the count on a plate is higher or lower than specified maximum and/or minimum bounds.

### Accepting a batch

While you are working in the Batch Designer, you can perform test measurements and experiment with and change the settings used to detect the colonies. Once you are satisfied with the design, the next step is to 'accept' the batch and switch aCOLyte3 to Measurement mode, which you then use to count the colonies on each plate, and record the results.

Once you have accepted the batch, some of the batch settings, such as whether you are detecting light or dark colonies, are fixed and cannot be changed. However, other settings, such as the sensitivity, small particle filter and colony splitter, can be changed in Measurement mode before you perform a count.

### Taking measurements

Once the batch has been accepted for Measurement mode, the process of counting the colonies is very straightforward: put a plate in the plate holder, press a button and move on to the next plate.

In most cases this is all you will need to do, but if there are any problems with difficult plates, you can make manual adjustments before carrying out the measurement. For example, for Pour Plate batches, you can add exclude regions, carry out a test measurement and optimize the colony detection settings, or perform a manual colony count.

After you have carried out a measurement, the results are recorded in a results table. You can edit the individual results in a number of ways. For example, you can add exclude regions and adjust the colony detection settings before carrying out an automatic colony recount, or you can add and remove colonies manually.

Any changes you make to a result are recorded in an audit history for that result.

Finally, once you have taken all the measurements, all you need to do to compile a report on the results is press a few buttons.

## Starting aCOLyte3

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**Note** You can control aCOLyte3 using the touch-sensitive screen, a mouse or keyboard shortcuts.

---

See the aCOLyte3 *Quick Guide* for how to set up the hardware ready for use.

To turn on the aCOLyte3 system:

1. Plug the flying lead from the power supply into the jack socket on the back of the aCOLyte3 system box.
2. Plug the power supply into a mains socket.
3. Plug in the USB lead.

## Getting started

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4. Plug in the software dongle.
5. Press the black button on the front of the instrument to switch the light on – the lighting can be cycled between using the lower lights, the upper light or both the upper and lower lights by repeatedly pressing this button.

To start the aCOLyte3 colony counting program:

1. Double-click on the aCOLyte3 icon on the Windows Desktop:



If user authentication is disabled (see *Managing aCOLyte3 users*, page 95), the program will open immediately and you will not need to follow the remaining steps in this procedure.

Otherwise, the **Please log on to aCOLyte3** dialog box will be displayed:



---

**Note** If user authentication is enabled, this dialog box is also displayed when you log out from aCOLyte3 or if you are logged out automatically by aCOLyte3 because you have not used it for a set period of time (5 minutes by default) – see *Logging out and closing aCOLyte3*, on the facing page, for more details.

---

2. Enter your Windows **Username** – the name you use to log on to Windows on this PC.
  3. Enter your Windows **Password** – the password you use to log on to Windows on this PC.
- 

**Notes** aCOLyte3 checks that the user name/password is a valid combination to log on to Windows on this PC, but does not check that it is the currently logged on user. This means that you can log on to aCOLyte3 while someone else is logged on to Windows on this PC.

---

4. Press **OK**.

The aCOLyte3 Application window will open. You can now either create a new 'batch' (see *Creating a completely new batch*, page 18) or open an existing batch if there is one (see *Opening and selecting batches*, page 61) and start counting colonies immediately.

### Logging out and closing aCOLyte3

To log out from aCOLyte3 (this only applies if user authentication is enabled – see *Managing aCOLyte3 users*, page 95):

Press the **Log out** button near the top right-hand corner of the aCOLyte3 application window:



The **Please log on to aCOLyte3** dialog box will be displayed.

---

**Note** You may also be logged out automatically and this dialog box displayed if you do not use aCOLyte3 for more than a set period of time (5 minutes by default) – see *System settings*, page 92, for how to set the timeout period.

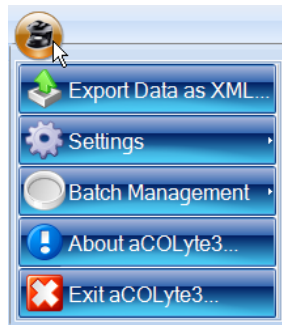
---

You (or someone else) can then:

- Log on again – see *Starting aCOLyte3*, page 3.
- Press **Close** to exit aCOLyte3.

Alternatively, to exit aCOLyte3:

1. Press the aCOLyte3 button at the top left-hand corner of the window to display the aCOLyte3 menu:



2. Press





# Capturing images

---

This chapter shows you how to capture and work with images. You will find out about:

- *The Image tab, below*
- *Loading plates into the instrument, on the next page*
- *Capturing an image, page 9*
- *Setting the exposure, page 9*
- *Zooming the image, page 10*
- *Exporting the image to a file, page 11*
- *Importing an image from a file, page 11*
- *Calibrating the image, page 12*
- *Viewing batch images, page 14.*

The 'view batch images' procedure is used to view images captured during the measurement process, so is only available in Measurement mode. All of the other procedures described in this chapter are used in both the Batch Designer and Measurement mode.

## The Image tab

To display the image controls:

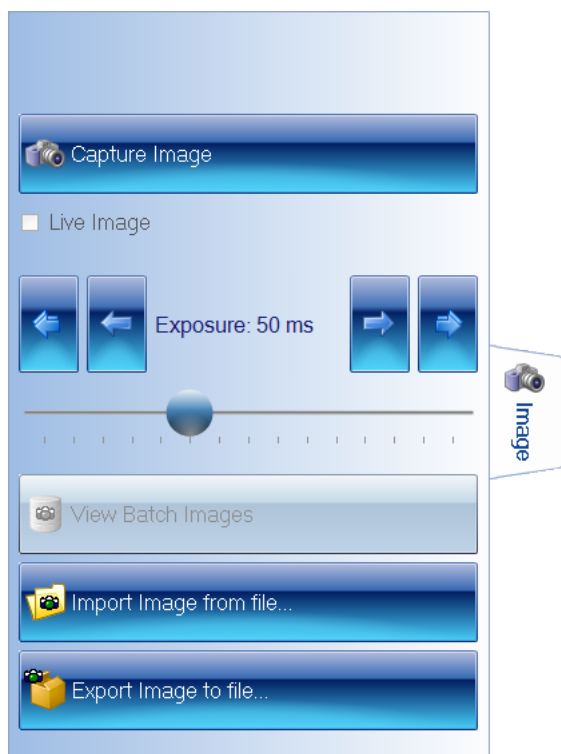
Press the **Image** tab:



## Capturing images

---

You use the controls on the **Image** tab to capture and work with images:



The View Batch Images button is enabled when you are working in Measurement mode (see *Using aCOlyte3 – overview*, page 2, for more information about the Batch Designer and Measurement mode).

For instructions on how to use the image controls, see the following sections:

- *Capturing an image*, on the facing page
- *Setting the exposure*, on the facing page
- *Exporting the image to a file*, page 11
- *Importing an image from a file*, page 11
- *Viewing batch images*, page 14.

See also:

- *Zooming the image*, page 10

## Loading plates into the instrument

To load a sample plate into the instrument:

1. Remove the curtain from around the sample stage – it is held in place by magnetic tape along its top edge.
2. If required, fit the small plate adapter in the plate recess.
3. Place the sample plate in the plate recess.

---

**Note** aCOlyte3 automatically displays a live image when you insert a new plate. However, if required, you can choose to display a live image manually by checking the **Live Image** box on the *The Image tab*, on the previous page.

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4. Re-attach the curtain around the sample stage.

### Capturing an image

To capture an image in aCOLyte3:

1. Press the **Image** tab



to display the image controls.

2. Press



The image will be captured and displayed in the aCOLyte3 window.

---

**Note** In fact, three images will be captured, with red, green and blue illumination respectively, and the results combined to form a color image – this procedure produces more accurate results than can be obtained using a color camera.

---

If the image is too light or too dark, you can adjust the exposure (see the next section, *Setting the exposure*) and capture the image again.

### Setting the exposure

---

**Note** You can adjust the exposure in both the Batch Designer and Measurement Mode.

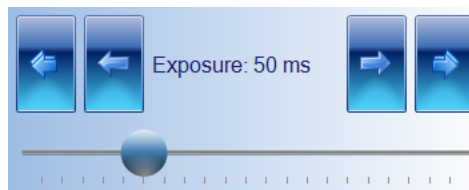
---

To set the exposure for capturing an image:

1. Press the **Image** tab



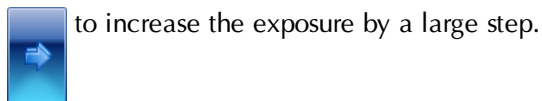
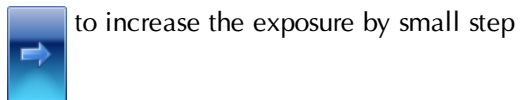
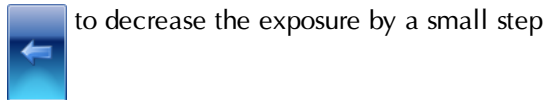
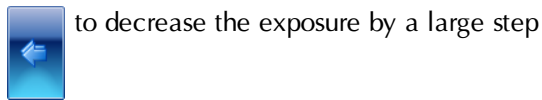
to display the image controls, including the exposure controls:



## Capturing images

---

- The current exposure time is shown between the arrow buttons. To change the exposure time, press:



Or:

Drag the handle in the exposure slider bar:



## Zooming the image

By default, the image is scaled to fit the image pane in the aCOLyte3 window.

To zoom the image:

Press



When it is zoomed, the button shows the scaling factor:



To zoom out:

Press



To return to fitting the image to the image pane:

Press





## Exporting the image to a file

To export the current captured image in the aCOLyte3 window to a file:

1. Press the **Image** tab



to display the image controls.

2. Press



to display the **Export Image** dialog box – this is a standard Windows 'Save As' dialog box.

3. Use the **Export Image** dialog box to:
  - select an image format from the **Save as type** drop-down list
  - select a folder to hold the image
  - enter a file name for the image.
4. Press **Save** to save the file.

### See also

*Viewing batch images*, page 14, for how to export previous images from a batch.

## Importing an image from a file

To import an image from a file into the aCOLyte3 window:

1. Press the **Image** tab



to display the image controls.

2. Press



to display the **Import Image** dialog box – this is a standard Windows 'Open' dialog box.

3. Use the **Import Image** dialog box to locate the folder holding the required image and select the image.
4. Press **Open** to open the file and load it into the aCOLyte3 window.

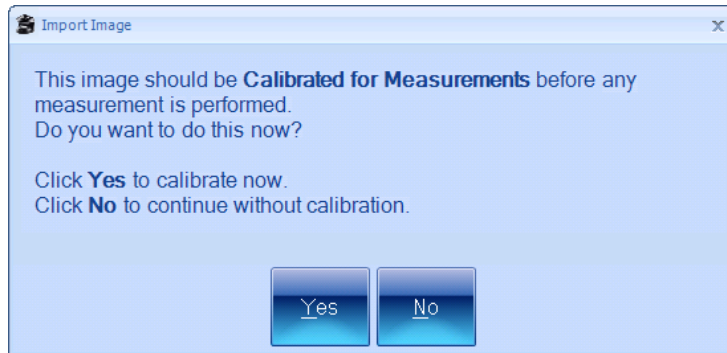
## Capturing images

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A warning dialog box will be displayed asking you to calibrate the image – see the next section, *Calibrating the image*, for details.

### Calibrating the image

When you import an image from a file (see *Importing an image from a file*, on the previous page) or try to perform a measurement from an uncalibrated imported image, a warning dialog box will be displayed asking if you want to calibrate the image, for example:



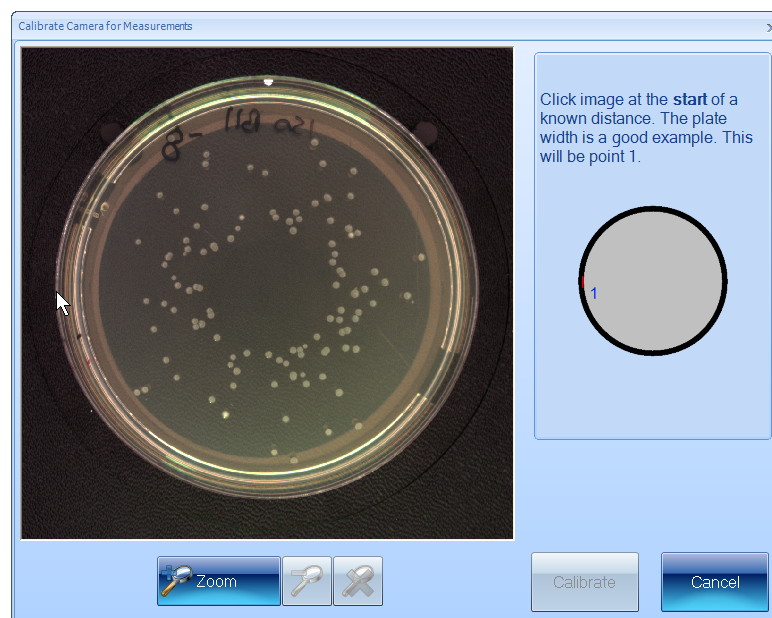
---

**Note** In order to create an accurate calibration you need to know the actual size of some feature in the image.

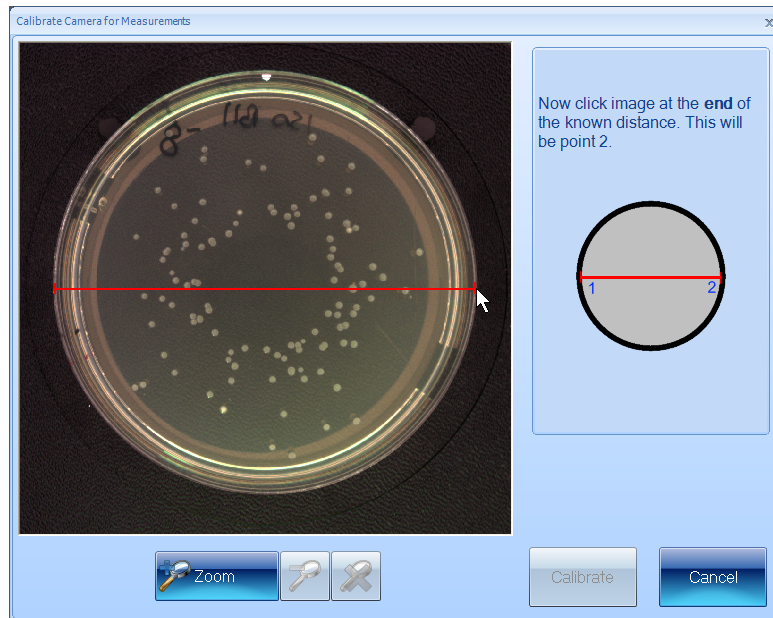
---

To calibrate the image:

1. Press **Yes** in the warning dialog box to display the **Calibrate Camera for Measurements** dialog box.
2. Choose some horizontal or vertical feature of known length in the image, such as the plate itself, and click on one end of it, for example:

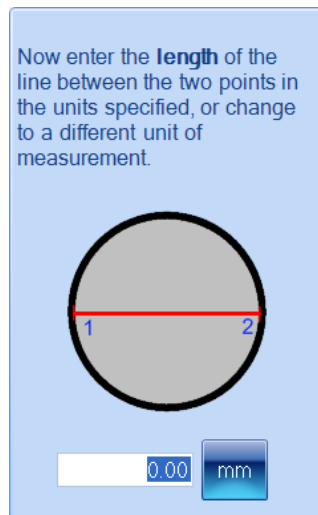


3. Move the pointer to the other end of the feature – as you do this, a line will be drawn back to the starting point, for example:

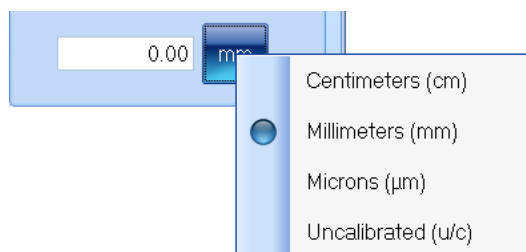


4. Click to mark the other end of the feature.

The calibration edit box and units button will be displayed:



5. To change the units:
  - a. Press the units button to display the units menu:



- b. Select the required units from the menu.

## Capturing images

6. Type the length of the known feature into the calibration edit box:



7. Press



to set the calibration and return to the main aCOLyte3 window.

## Viewing batch images

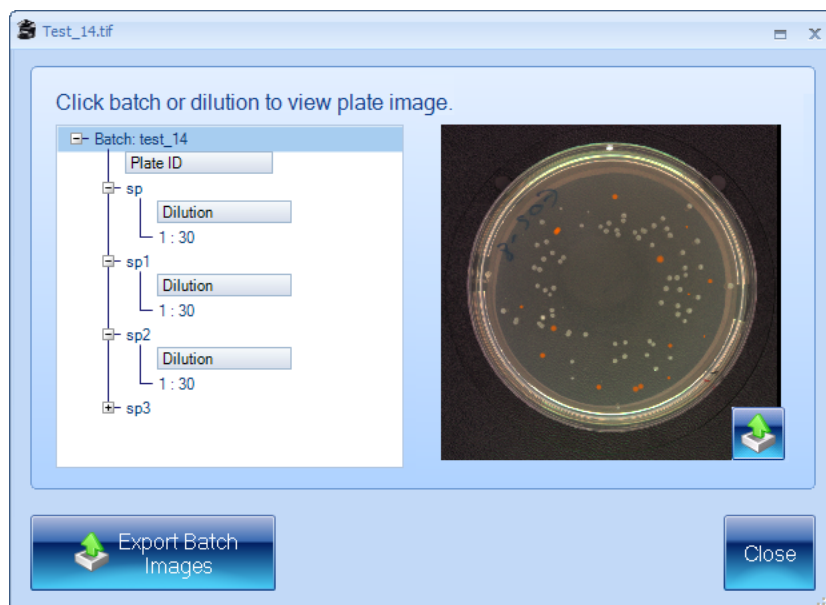
By default (see *System settings*, page 92), when you create a new batch, the image used to set it up (see *Setting up batches*, page 24) is saved with the batch, and then, after you have accepted the batch design, each time you take a measurement in Measurement mode, the captured image is also saved with the result in the batch.

To view the batch setup image and images used for measurements:

1. Press



to display a dialog box:



The box on the left-hand side lists the batch images, beginning with the batch setup image followed by the measurement images listed in order of their plate identifiers – expanding the measurement image items (as in the picture) shows the dilutions.

2. To view a measurement image, click on the plate identifier.

To view the batch setup image, click on the batch identifier (the top line).

3. To export all the batch images:

a. Press



to display the **Browse for Folder** dialog box.

b. Select the folder you want to export the images to – if required, press **Make New Folder** to add a new folder to the selected folder.

c. Press **OK** to save the batch images to the selected folder.

The batch setup image will be saved using the name of the batch; the measurement images will be saved using the plate identifier.

An xml file containing information about the images will also be saved in the folder.

4. To export a single batch image:

a. Select the required image – see Step 2.

b. Press



to display a Standard Windows **Save As** dialog box.

c. Use the **Save As** dialog box to select a folder and enter a filename for saving the image.

d. Press **Save** to save the selected image.

5. Press **Close** to close the dialog box and return to the main aCOLyte3 window.



# Creating a new batch

---

This chapter shows you how to create and set up a new batch for counting colonies on plates. It also describes a number of batch management procedures for use once you have created the batch.

The process of creating a new batch is divided into two main stages, which are covered in the following main sections:

- *Creating the batch* – see the next section

This section shows you how you can create:

- A completely new batch from scratch – see *Creating a completely new batch*, on the next page.
- A new batch based on an existing batch or an exported batch file – see *Creating a new batch based on an existing batch or an exported batch file*, page 21.
- *Setting up batches*, page 24.

This section describes:

- *Giving the batch a name*, page 25
- *The Classification tab*, page 25, for specifying how colonies are detected.
- *The Configuration tab*, page 35, for setting up how plate identifiers will be assigned to plates when they are measured.
- *The Measure tab*, page 47, including:
  - *Plate ID*, page 48
  - *Dilution*, page 48
  - *Setting count restrictions*, page 52
  - *Test Measure Plate*, page 53.
- *The Results tab*, page 54, for choosing what to display in the Results table.

Once you have completed all the settings, you will need to accept the batch so that you can use it for measuring plates – see *Accepting the batch design*, page 56.

---

**Note** The sections described above show you how to use the Batch Designer, but many of the operations covered may also be carried out when you are working in Measurement mode.

---

The final sections of this chapter describe a number of batch management operations – for details, see:

- *Exporting batch details*, page 57
- *Deleting batches*, page 57.

## Creating the batch

Before you can start counting colonies on plates you need to set up and create a 'batch' to define how the counts should be carried out, and to hold the results.

## Creating a new batch

---

You can create:

- A completely new batch from scratch – see *Creating a completely new batch*, below.
- A new batch based on an existing batch or an exported batch file – see *Creating a new batch based on an existing batch or an exported batch file*, page 21.

---

**Note** Once you have created the new batch, you will still need to set it up and test it before you can start using it to count colonies – see *Setting up batches*, page 24.

---

## Creating a completely new batch

---

**Note** See *Creating a new batch based on an existing batch or an exported batch file*, page 21, for how to create a batch with the same settings as an existing or exported batch.

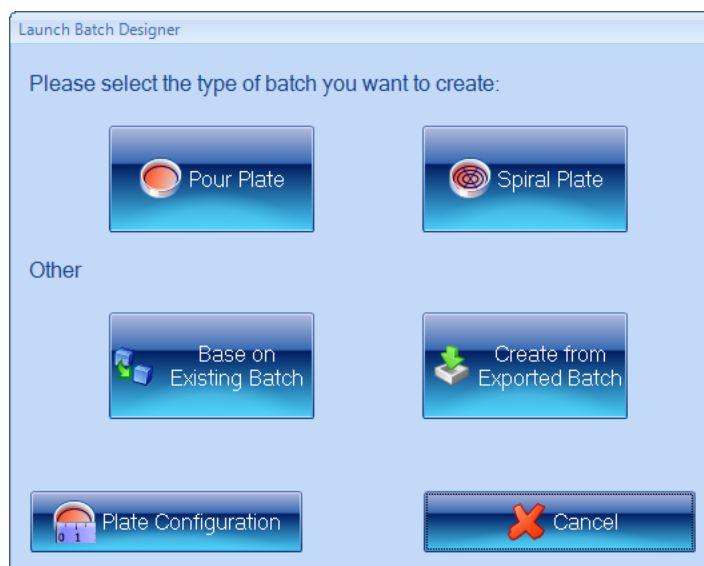
---

To create a completely new batch:

1. Press



to display the **Launch Batch Designer** dialog box:

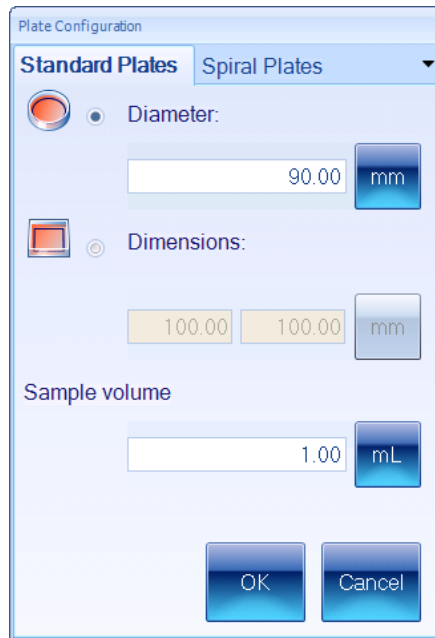


2. To set the plate configuration:
  - a. Press





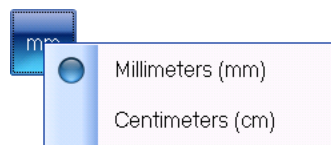


to display the **Plate Configuration** dialog box:



If you are going to use the batch for spiral plates, go to Step c; otherwise, go to Step b.

- b. For pour plates:
  - i. Click on the **Standard Plates** tab if it is not already selected – see picture in Step a.
  - ii. Press the appropriate radio button to specify whether the plates you are using are:
    -  = circular
    - or
    -  = rectangular.
  - iii. For a circular plate, enter the **Diameter** of the plate; for a rectangular plate, enter the **Dimensions**.
  - iv. To choose the units for the **Diameter** or **Dimensions**, press the corresponding units button to display the units menu



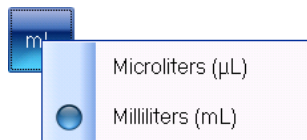
and select the required unit.

- v. Enter the **Sample volume** you will be using.

## Creating a new batch

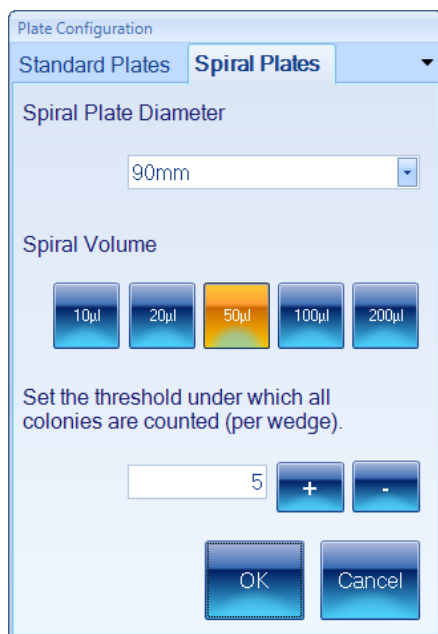
---

- vi. To choose the units for the **Sample volume**, press the units button to display the units menu



and select the required unit.

- vii. Press **OK** to confirm the settings, close the dialog box and return to the **Launch Batch Designer** dialog box – go to Step 3.
- c. For spiral plates:
    - i. Click on the **Spiral Plates** tab to display the **Spiral Plates** page:



- ii. Select the **Spiral Plate Diameter** you will be using – you can select **90mm** or **Oxoid 70mm**.
- iii. Click on the button corresponding to the **Spiral Volume** you will be using.
- iv. If you are using a two-sector counting frame and the number of colonies counted within the first sector is less than a set threshold, the whole frame will be used instead.

To **Set the threshold under which all colonies will be counted**, type in a value directly or press the **+** or **-** button to increase or decrease the current value.
- v. Press **OK** to confirm the settings, close the dialog box and return to the **Launch Batch Designer** dialog box.

---

**Note** You will not be able to change the settings made in the **Plate Configuration** dialog box after you have created the batch in the next step.

---

3. To create the new batch, press the button to create the new **Pour Plate** or **Spiral Plate** batch.

The Batch Designer will be launched for the selected plate type so that you can complete the process of setting up the new batch – see *Setting up batches*, page 24, for details.

### Creating a new batch based on an existing batch or an exported batch file

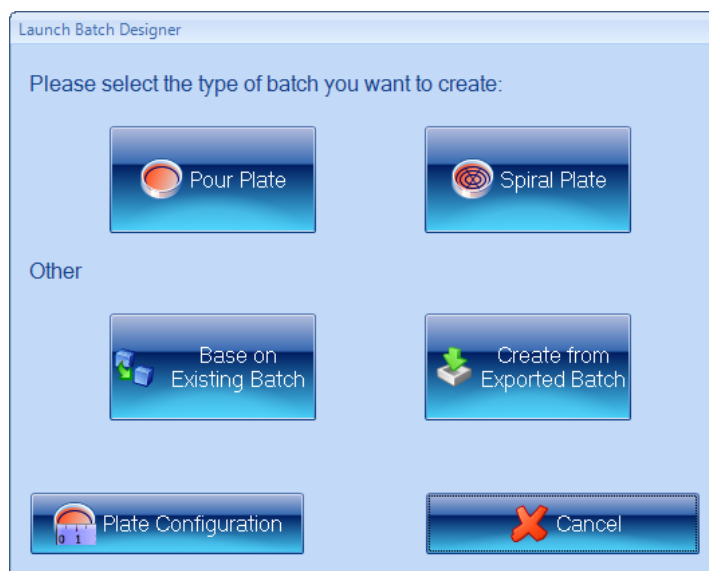
**Note** See *Creating a completely new batch*, page 18, for how to create a new batch from scratch.

To create a new batch based on an existing batch or an exported batch file:

1. Press



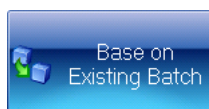
to display the **Launch Batch Designer** dialog box:



Go to Step 2 if you want to create a new batch from an existing batch; go to Step 3 if you want to create a new batch from an exported batch file.

**Note** The plate configuration will be inherited from the existing batch and any settings you make using the **Plate Configuration** button will be ignored.

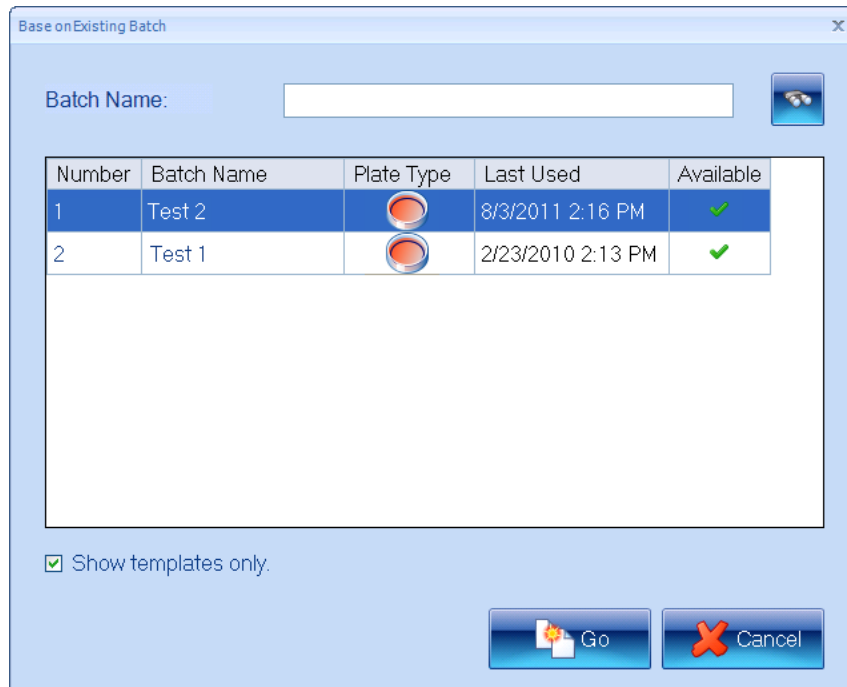
2. If you want the new batch to have the same settings as an existing batch:
  - a. Press



## Creating a new batch

---

to display the **Base on Existing Batch** dialog box:



---

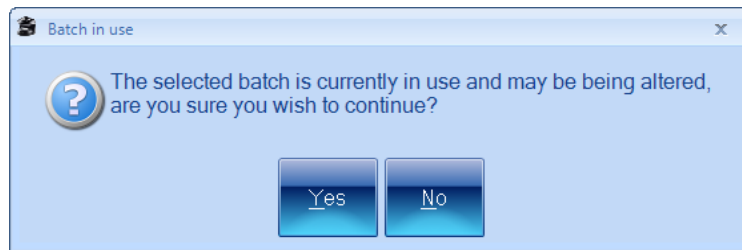
**Note** Any batches that are currently open will be marked with in the **Available** column. You will not be prevented from choosing to base the new batch on one of these, but you will be warned when you confirm the selection – see Step f.

---

- b. Select **Show templates only** to hide any batches in the list that were based on an existing batch or an exported batch file.
  - c. If there are a large number of existing batches, the batch you want to use as a basis for the new batch may not be shown in the list. To search for an existing batch:
    - i. Type the name, or a part of the name, of the required batch into the **Batch Name** box.
    - ii. Press
- Only batches with names that contain the text you entered will be listed.
- d. Select the required batch in the list.
  - e. Press



- f. If you chose an existing batch that is currently in use, you will be warned:



Press **Yes** to create the new batch anyway; press **No** to close the warning dialog box and return to Step c to choose another batch.

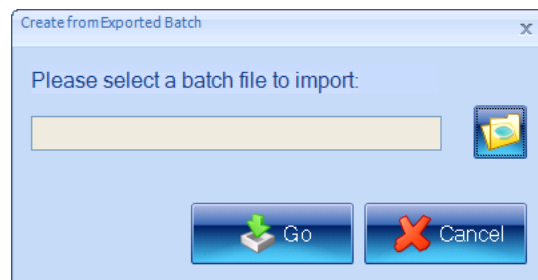
The new batch will be created; see the paragraph following Step 3 for what to do next.

3. If you want the new batch to have the same settings as an exported batch (see *Exporting batch details*, page 57, for how to export batches):

- a. Press



to display the **Create from Exported Batch** dialog box:



- b. Press



to display the **Please select a batch file to import** dialog box – this is a standard Windows **Open** dialog box.

- c. Use the dialog box to locate and select the required exported batch file.  
 d. Press **Open** to select the file and return to the **Create from Exported Batch** dialog box.  
 e. Press



to create the new batch.

The new batch will be created and loaded into the Batch Designer with the same settings as the selected existing batch or exported batch file. You will be able to edit some of these settings in the Batch Designer – see *Setting up batches*, on the next page, for how to customize the batch settings in the Batch Designer.

## Creating a new batch

---

**Note** You will not be able to edit the settings made in the **Plate Configuration** dialog box when the original batch was created – see *Creating a completely new batch*, page 18, for details of the settings made in the **Plate Configuration** dialog box.

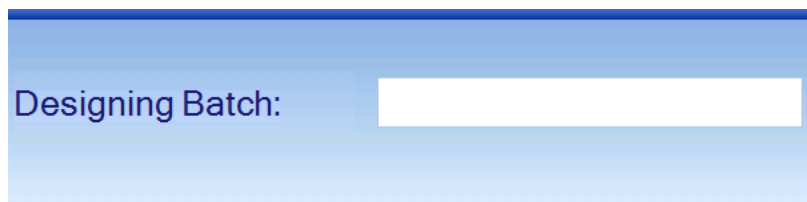
---

## Setting up batches

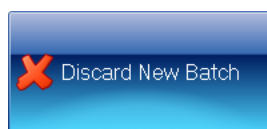
Once you have created a new batch (see *Creating the batch*, page 17), it will be loaded into the Batch Designer for final set up and testing before you use it to carry out and record measurements.

While the Batch Designer is active:

- The message area at the bottom of the aCOLyte3 window contains an edit box allowing you to enter a name for the batch (see *Giving the batch a name*, on the facing page):

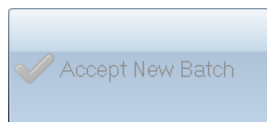


- The **Discard New Batch** button appears near the top left corner of the aCOLyte3 window:



Press **Discard New Batch** to abort the new batch – you will be asked to confirm that you want to do this.

- The **Accept New Batch** button appears near the top left corner of the aCOLyte3 window. Initially, the button will be disabled:



When you have completed the steps *required* to set up the batch, the button will become enabled:



See *Accepting the batch design*, page 56, for further details.

You will not be able to accept a batch design until you have given it a name – see the next section *Giving the batch a name*.

The other settings you will need to make when you are creating a completely new batch, or the settings you can edit if the new batch is based on an existing one, are grouped on a series of tabs – see the following sections for details:

- *The Classification tab*, on the facing page

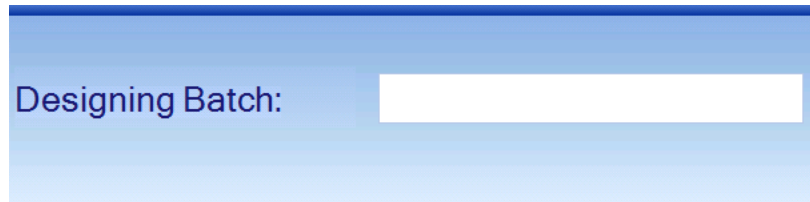
- *The Measure tab*, page 47
- *The Configuration tab*, page 35
- *The Results tab*, page 54.

### Giving the batch a name

After you have created a new batch (see *Creating the batch*, page 17), you must give it a name before you can accept the design (see *Accepting the batch design*, page 56).

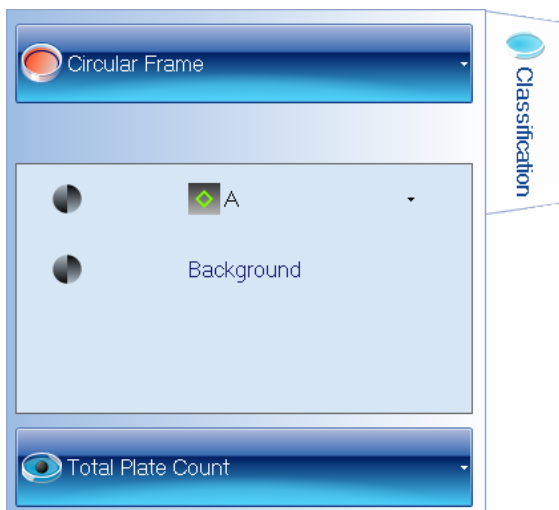
To give a name to a batch:

Type the name into the **Designing Batch** box at the bottom of the **Batch Designer** screen:



See *Setting up batches*, on the previous page, for a list of other steps in setting up a new batch.

### The Classification tab



To display the classification controls in the Batch Designer:

Press the **Classification** tab:



## Creating a new batch

---

In order to use the **Classification** tab, you will need a sample image to work with:

1. Insert a typical plate from the batch into aCOLyte3 – see *Loading plates into the instrument*, page 8.
2. Capture the image – see *Capturing an image*, page 9.

The **Classification** tab in the Batch Designer for counting colonies allows you to:

- set the frame – see *Frame*, below
- configure a simple total plate count – see *Total Plate Count*, page 30
- set the properties of the markers used to identify detected colonies on the image – see *The Classification panel*, page 33

---

**Note** By default, if you base a new batch on an existing batch, it will use the same total plate count settings as the existing batch (though you can change them if required). If you are creating a completely new batch, you will not be able to accept it for taking measurements until you have defined a total plate count.

---

## Frame

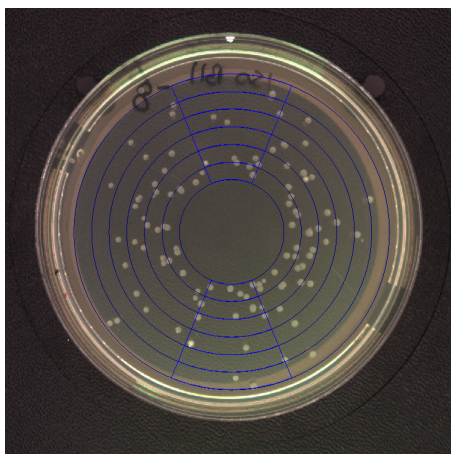
The following sections tell you about:

- *Choosing the type of frame to use*
- *Adjusting the position and size of frames*, page 28.

### **Choosing the type of frame to use**

The ‘frame’ defines the area within which colonies will be counted. The choice of frames depends on the type of batch:

- **Pour Plate:** the frame can be circular, rectangular or a single sector of a circle occupying 1/10, 1/8, 1/4 or 1/2 of the total area.
- **Spiral Plate:**
  - Whole Frame – Whole Frame: the frame is circular
  - Whole Frame – Annulus Frame: the frame consists of two concentric circles
  - Two Sector: a series of concentric circles, with two opposite highlighted sectors. For example:





To change the type of frame used to define the region for counting colonies:

1. Press the frame button to display the frame menu.
2. Choose the required option – the contents of the menu depend on the type of plate and are described in the following sections:
  - *Pour Plate batches*, below
  - *Spiral Plate batches*, below.

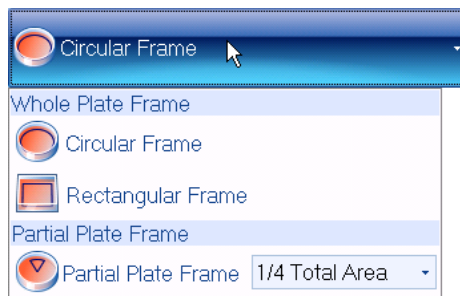
---

**Note** You can change the type of frame in the Batch Designer or Measurement mode.

---

### Pour Plate batches

You can choose **Circular Frame** or **Rectangular Frame**, or, to use a frame covering a single sector only, choose **1/2**, **1/4**, **1/8** or **1/10** from the **Partial Plate Frame** submenu:



Partial plate frames can be useful when you have plates that are heavily loaded with bacteria, or if there is some problem with a plate and you just want to read results from an unaffected part. This means that you are more likely to want to choose a partial plate frame for an individual plate while you are working in Measurement mode than for all the plates in a batch when you are creating it in the Batch Designer.

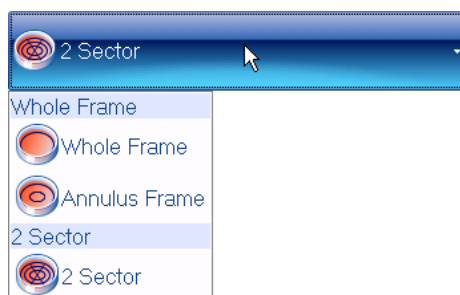
---

**Note** You can also avoid counting in problematic areas of individual plates by placing Exclude regions over them – see *Exclude regions*, page 69.

---

### Spiral Plate batches

You can choose **Whole Frame**, **Annulus Frame** or **2 Sector**:




---

**Notes** As a general rule, if there has only been a small amount of colony growth, you should use one of the **Whole Frame** options (use **Annulus Frame** if the spiral plater does not reach the center of the plate; use **Whole Frame** if it does reach the center), otherwise, use **2 Sector**.

If you choose **2 Sector** and the number of colonies counted in the first sector falls below the **threshold under which all colonies are counted** (see *Creating a completely new batch*, page 18, or ), aCOLyte3 will count the whole frame.

---

### ***Adjusting the position and size of frames***

The following sections describe how to adjust the position and size of frames:

- *Moving frames*, below
- *Resizing frames*, below
- *Rotating frames*, on the facing page.

---

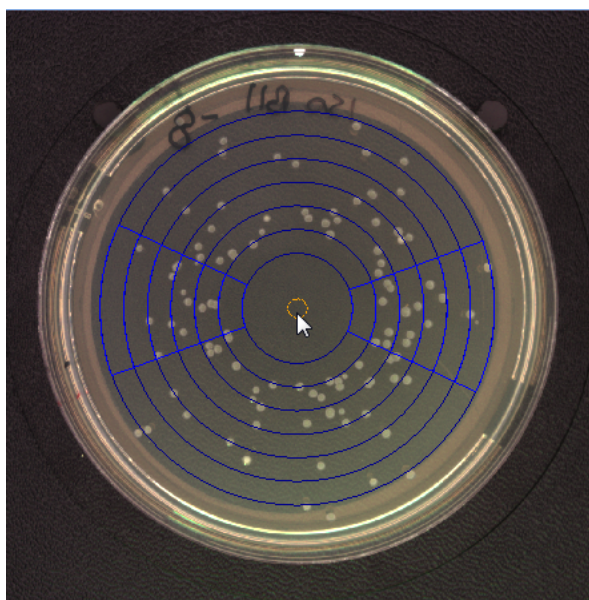
**Note** You should load a plate from the batch into the instrument and capture an image before adjusting the frame – see *Loading plates into the instrument*, page 8, and *Capturing an image*, page 9.

---

### **Moving frames**

To move a frame:

1. Move the pointer near to the center of the frame – an orange circular drag handle will appear in the center of the frame. For example:



2. Drag the frame and drop it in the required position.

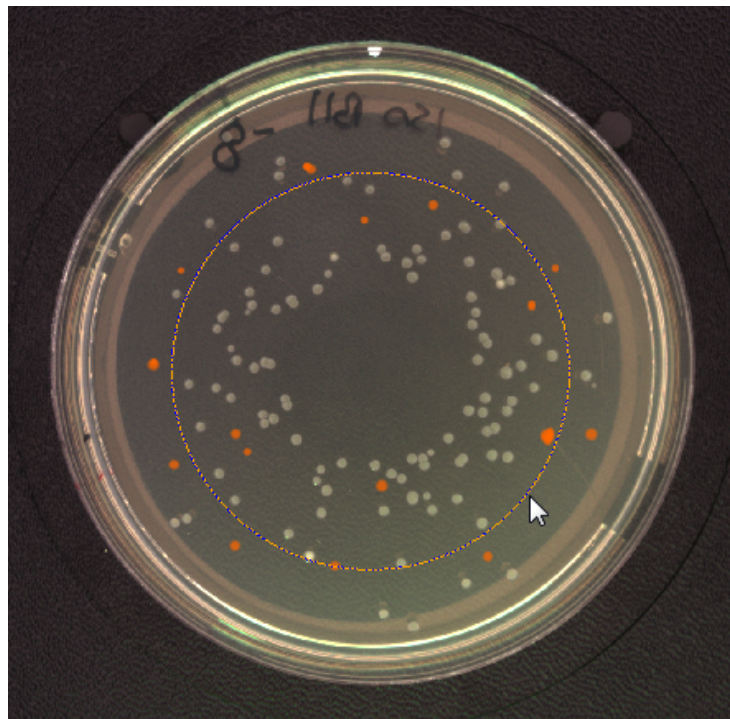
### **Resizing frames**

Generally, frames should be made as large as possible within the area of the plate but avoiding the very edge (as this may produce spurious results) and allowing some margin for variation between plates. You can adjust the size for individual plates in Measurement mode, but it will save time if you can use a single setting for all the plates in a batch.

To resize a frame:

1. Move the pointer near to the frame boundary (for Partial Plate frames, the frame boundary is the circle of which the frame forms a sector).

The frame boundary will turn orange – showing that you can adjust its size. For example:



2. Drag the frame boundary to resize/reshape the frame.

For rectangular frames, drag the top or bottom side to adjust the height; drag the left or right side to adjust the width; drag a corner if you want the frame to maintain the same shape (aspect ratio) when you adjust the size.

### Rotating frames

You can rotate 2-Sector Spiral Plate frames and rectangular and Partial Plate Pour Plate frames.

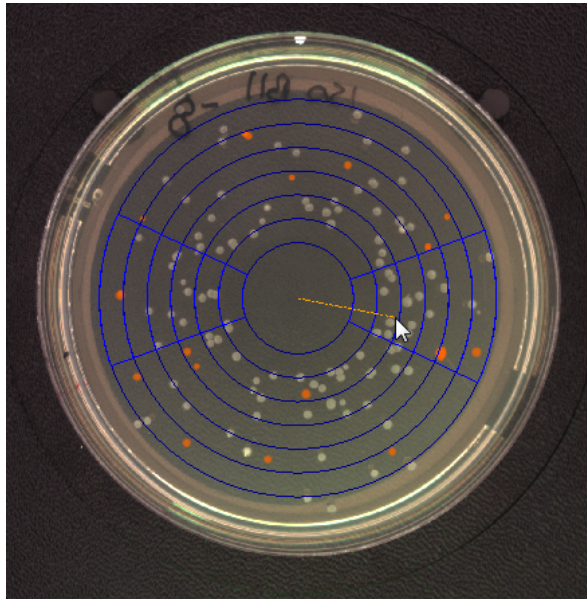
To rotate a frame:

1. Move the pointer to a point about half way between the center and edge of the frame.

## Creating a new batch

---

An orange line will appear joining the pointer to the center. For example:



2. Drag the line around the center to rotate the frame.

## Total Plate Count

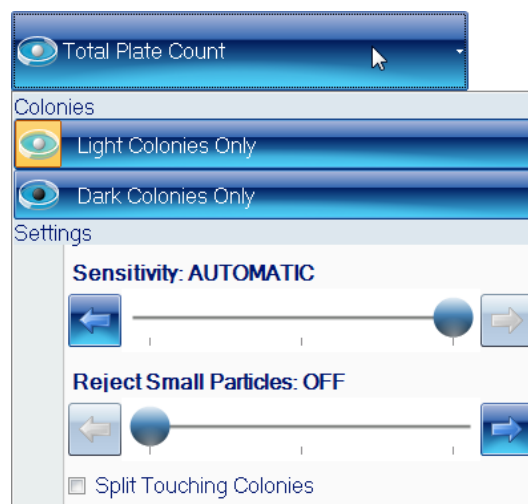
You can use the **Total Plate Count** button to set simple criteria for detecting and counting the total number of colonies on a plate.

To set the **Total Plate Count** criteria:

1. Press the **Classification** tab in the Batch Designer – see *The Classification tab*, page 25.
2. To specify whether the colonies to be counted are lighter than or darker than the background:
  - a. Press:



to display the **Total Plate Count** controls:



**Note** If there is no current image, an image will be automatically captured when you press the **Total Plate Count** button. If the image shown in the viewer is too light or too dark, press the **Image** tab and adjust the exposure as required (see *Setting the exposure*, page 9), then go back to Step 1.

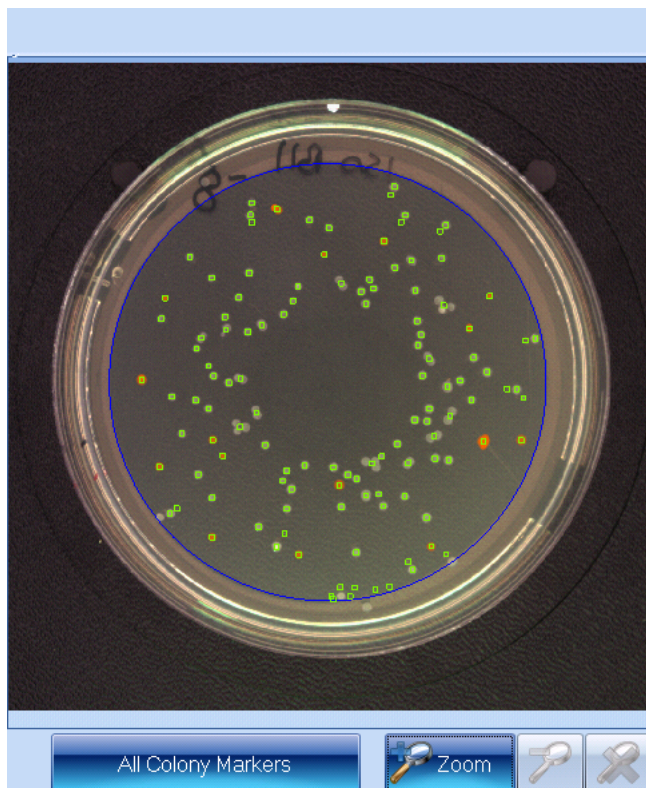
- b. Press the appropriate button to choose whether to count:
- **Light Colonies Only** – the number of colonies lighter than the background
  - **Dark Colonies Only** – the number of colonies darker than the background.

The **Total Plate Count** controls will be hidden and the icon on the **Total Plate Count** will show the selected option:



or

- c. If the button below the image is labeled **No Colony Markers**, press the button and select **All Colony Markers** to display the detected colonies on the image:



For more detailed information about the precise areas identified as colonies, you can also select:

- **All Colony Outlines:** the boundary of each detected colony is marked in the color of the symbol representing the type of colony
- **All Colony Regions:** the area of each detected colony is marked in the color of the symbol representing the type of colony.

If required, you can zoom the image to check the colony detection – see *Zooming the image*, page 10.

## Creating a new batch

---

If required, you can change the symbol and color used to mark the colonies – for details, see the next section, *The Classification panel*.

3. To adjust the settings used when counting colonies (if the button below the image is labeled **All Colony Markers**, **All Colony Outlines** or **All Colony Regions** the detected colonies will be shown on the image when you make the adjustments):
  - a. Press:



to display the **Total Plate Count** controls again – see picture in Step 1.

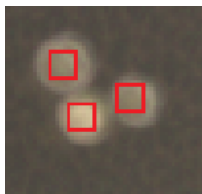
- b. Click on the **Split Touching Colonies** check button to specify whether to use the colony splitter for colonies or not.

For example:

- **Split Touching Colonies** unchecked:



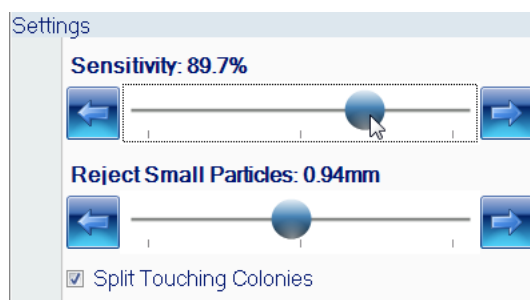
- **Split Touching Colonies** checked:



- c. In most cases, the automatic sensitivity setting will give excellent results, but if the colonies are not being detected correctly, you can experiment by adjusting the sensitivity manually until you get an optimal result.

To adjust the detection sensitivity:

Drag the **Sensitivity** slider; click the arrow buttons at the ends of the slider to adjust the slider by a single step; or press anywhere on the slider bar to set the slider to that position – as you adjust the setting, the control shows the sensitivity set:

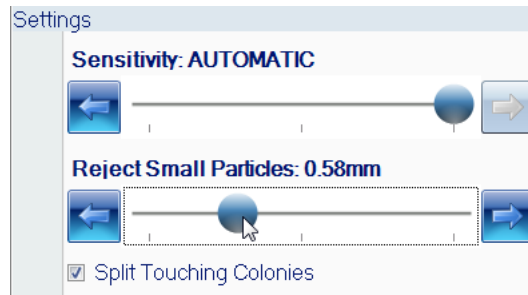


To set the **Sensitivity** back to **AUTOMATIC**, drag the slider back to the right-hand end – in fact, this corresponds to a manual sensitivity setting at the middle of the scale.



- d. To set a lower limit to the size colonies must be in order to be counted (you can use this, for example, to prevent small particles of debris being counted as colonies):

Drag the **Reject Small Particles** slider; click the arrow buttons at the ends of the slider to adjust the slider by a single step; or press anywhere on the slider bar to set the slider to that position – as you adjust the setting, the control shows the minimum colony size set:



To switch small particle filtering off, drag the slider to the left-hand end.

**Note** The **Sensitivity**, **Reject Small Particles** and **Split Touching Colonies** settings you make here will be used to perform the initial counts for plates when you are working in Measurement mode. However, you will be able to vary these settings for individual plates by carrying out a test count in Measurement mode (see *Test measurement and adjusting settings*, page 72), or by editing the result after you have carried out a count (see *Rejecting small particles in a result*, page 84, *Changing the Sensitivity setting for a result*, page 85, and *Splitting colonies in a result*, page 85).

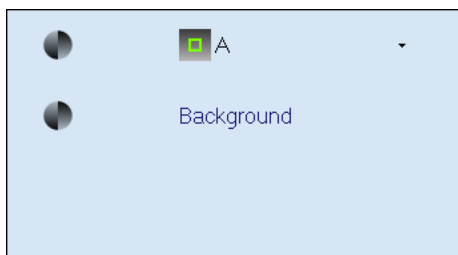
4. Press



to hide the **Total Plate Count** controls.

## The Classification panel

The Classification panel appears below the frame button in the classification tab. After you have selected **Total Plate Count** (see *Total Plate Count*, page 30), it shows a key for the markers used to identify the detected colonies on the image:



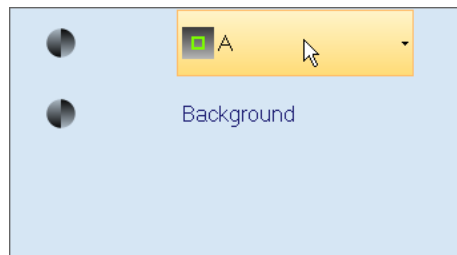
The Classification panel allows you to change the colony marker and color.

## Creating a new batch

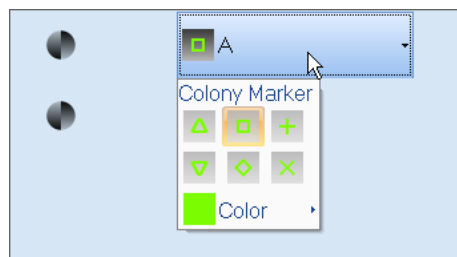
---

To change the colony marker or color:

1. Move the pointer over the colony item to display the colony button:



2. Press the colony button to display the marker controls:



3. Either:

Select the shape of colony marker you want to use from the marker controls.

Or:

- a. Press **Color** to display the **Standard Colors** control:



- b. Click on the required color in the **Standard Colors** control.



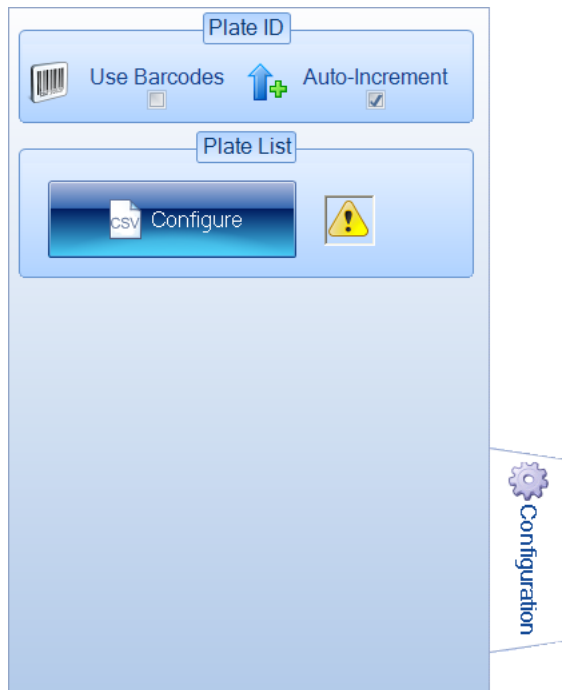
---

## The Configuration tab

---

**Note** The **Configuration** tab does not appear in Measurement mode.

---



---

**Note** The **Use Barcodes** check box is disabled if there is no barcode reader attached to the PC.

---

To display the configuration controls:

Press the **Configuration** tab:



The **Configuration** tab in the Batch Designer allows you to specify how plate identifiers and, where relevant, dilutions should be assigned to plates in measurement mode.

## Creating a new batch

---

If the batch you are creating is based on another batch, the **Configuration** tab shows the settings selected in the parent batch:



The green check marks show the settings selected in the parent batch – if the parent batch uses manually entered parent plate identifiers, the **Parent is using auto-increment** will have a red cross next to it, but its check box will be enabled.

To use the same configuration tab settings as in the parent batch:

Leave the **Parent Batch** check boxes unchanged.

---

**Note** If the **Parent is using a plate list**, you will need to create a new plate list for the new batch – see *Selecting auto-incrementing plate identifiers*, page 39, for instructions.

---

If you do not want to use the same configuration tab settings as in the parent batch:

Uncheck the **Parent Batch** check box(es) for the settings you do not want to use – see the references below for how to set independent configuration tab settings for the current batch.

If you are creating a new batch from scratch, or have chosen not to use the parent batch settings, you can choose to use:

- **Manual plate identifiers and dilutions**

You will need to enter an identifier manually for each batch and, if required, manually change the dilution.

See *Manually entered plate identifiers*, on the facing page, for how to choose this option.

- **Auto-incrementing plate identifiers and manual dilutions**

Automatic identifiers will be created for each plate as it is measured, and, if relevant, you can manually change the dilution.

See *Selecting auto-incrementing plate identifiers*, page 39, for details.

- **A plate list of plate identifiers and, optionally, dilutions**

Each identifier (and dilution) in the list is assigned to each of the plates in turn as they are measured. You can enter a plate list by hand or import it from a file.

See *Using an external source for plate identifiers*, page 42, for details.

- **A barcode reader**

The barcode reader can be used to read identifiers and, depending on the setup, dilutions directly from a barcode on the plate to enter identifiers/dilutions manually or in combination with a plate list.

See *Using a barcode reader for plate identifiers*, page 41, for details.

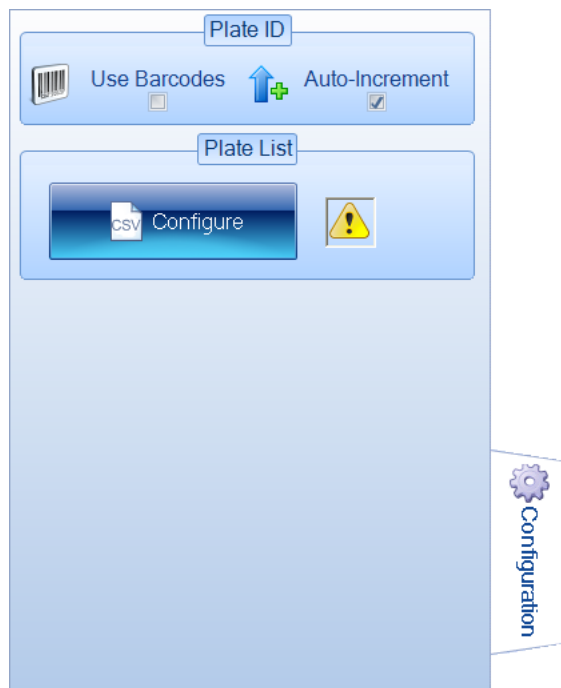
### Manually entered plate identifiers

To set up a batch to use manually entered plate identifiers and, where relevant, dilutions:

1. Press



to display the **Configuration** tab:



2. If the new batch is based on an existing batch, there will be a set of **Parent Batch** controls below the **Plate List** controls. If the parent batch did not use manually entered plate identifiers, uncheck the **Parent Batch** check box(es) – see *The Configuration tab*, page 35, for details.

## Creating a new batch

---

3. If the icon in the **Plate List** box is



go straight to Step 4.

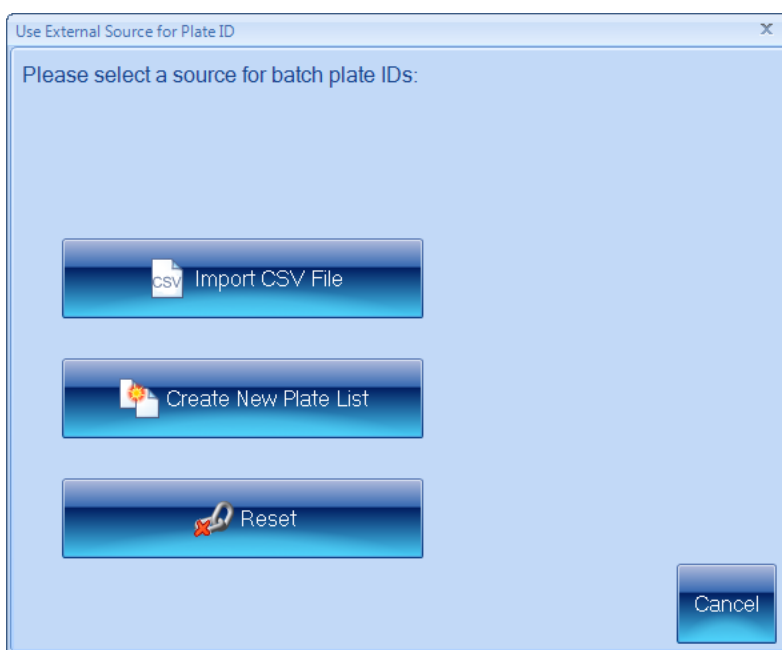
Otherwise, an external source of plate identifiers has been selected for the batch and the **Auto-Increment** control will be disabled.

To enable the **Auto-Increment** control and deselect the external source:

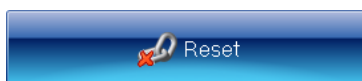
- a. Press



to display the **Use External Source for Plate ID** dialog box:



- b. Press



4. If **Auto-increment** is selected in the **Plate ID** box, click the check box to deselect it.
5. If you want to read the manually entered plate identifiers (and dilutions, depending on your setup) from a barcode on the plate, select **Use Barcodes** in the **Plate ID** box – see *Using a barcode reader for plate identifiers*, page 41, for details.

## Selecting auto-incrementing plate identifiers

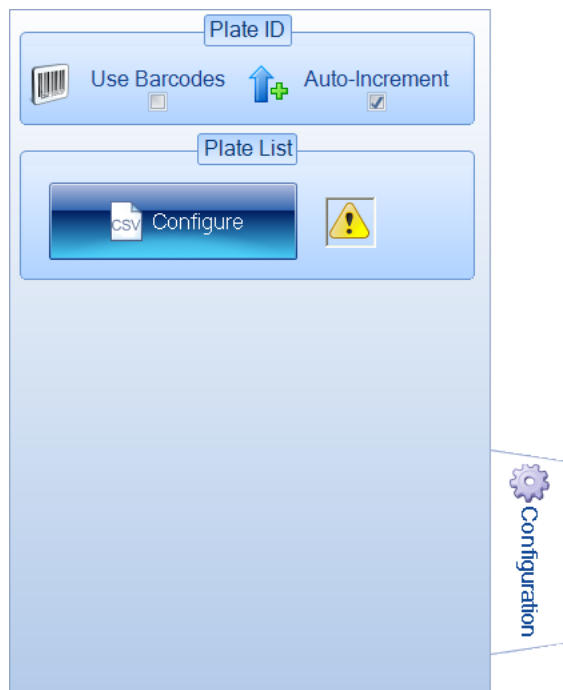
**Note** This option is selected by default when you create a new batch from scratch.

To use auto-incrementing plate identifiers:

1. Press



to display the **Configuration** tab:



2. If the new batch is based on an existing batch, there will be a set of **Parent Batch** controls below the **Plate List** controls. If the parent batch did not use manually entered plate identifiers, uncheck the **Parent Batch** check box(es) – see *The Configuration tab*, page 35, for details.
3. If the icon in the **Plate List** box is



go straight to Step 4.

Otherwise, an external source of plate identifiers has been selected for the batch and the **Auto-Increment** control will be disabled.

## Creating a new batch

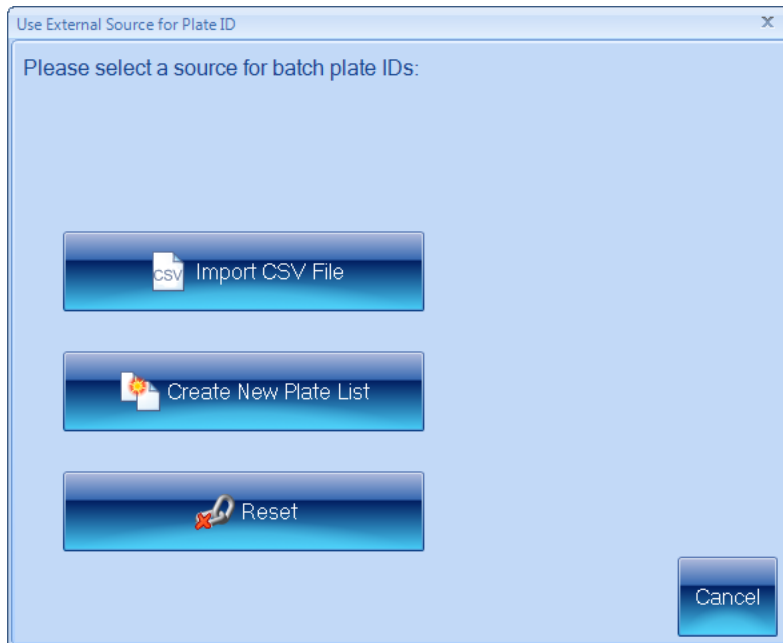
---

To enable the **Auto-Increment** control and deselect the external source:

- a. Press



to display the **Use External Source for Plate ID** dialog box:



- b. Press



4. If **Auto-increment** is not selected in the **Plate ID** box, click the check box to select it.

5. Press the **Measure** tab



to display the measure controls (see *The Measure tab*, page 47).

6. In the **Plate ID** box, type in text to act as a basis for the identifier.

If the text includes a sequence of digits, these will be used as the starting point for the auto-incrementing; if it includes more than one sequence, the rightmost sequence will be used as the starting point; if there are no digits in the text, auto-incrementing digits will be added to the end of the text. For example:

Template	First identifier	Second identifier	Third identifier
abc	abc1	abc2	abc3
abc18	abc19	abc20	abc21
abc18def	abc19def	abc20def	abc21def
66abc18def	66abc19def	66abc20def	66abc21def

## Using a barcode reader for plate identifiers

**Note** Depending on how the barcode reader is programmed, the barcode reader may or may not be able to read dilution information.

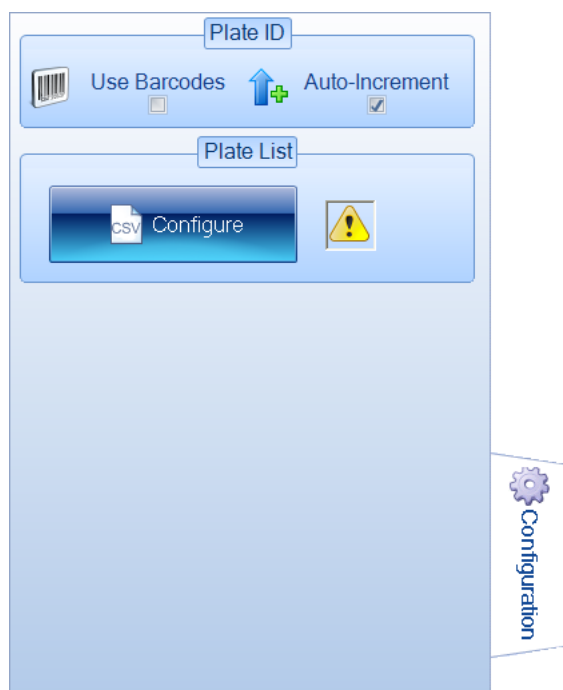
When you are working in Measurement mode, you can use a barcode reader to read the identifier/dilution from the plate in combination with an external source of plate identifiers (see *Using an external source for plate identifiers*, on the next page) or to input manual identifiers (see *Manually entered plate identifiers*, page 64).

If you intend to use a barcode reader to read plate identifiers (and dilutions where relevant – see the note above):

1. Make sure there is a barcode reader attached to the PC – see the note in Step 2.
2. Press



to display the **Configuration** tab:



**Note** The **Use Barcodes** check box is disabled if there is no barcode reader attached to the PC.

3. Select **Use Barcodes**.

See *Using a barcode reader for identifiers and dilutions*, page 68, for more on using the barcode reader to input plate identifiers/dilutions.

## Creating a new batch

---

### Using an external source for plate identifiers

You can use the following external sources for plate identifiers:

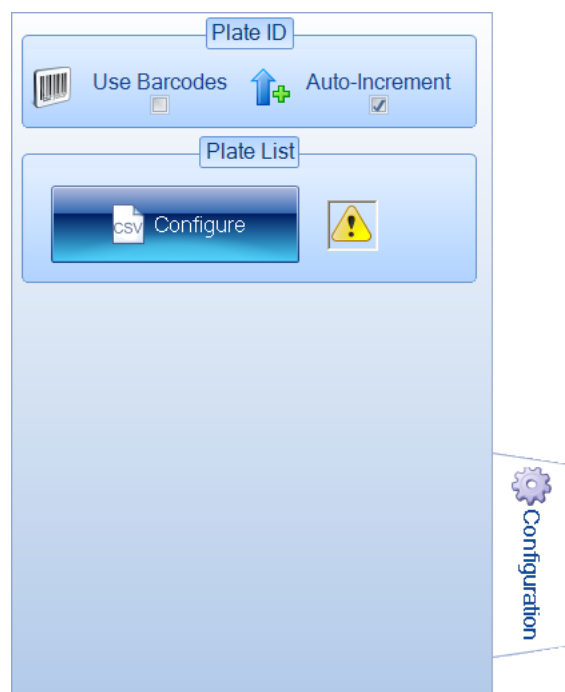
- a CSV file
- a list created in aCOLyte3.

To specify an external source for plate identifiers:

1. Press



to display the **Configuration** tab:

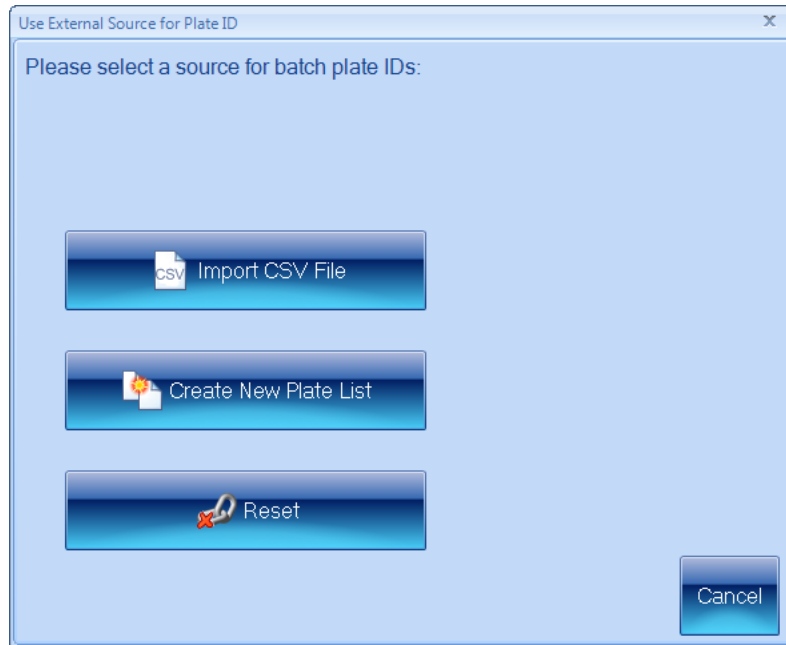


2. Press





to display the **Use External Source for Plate ID** dialog box:



3. Press the button for the required option.

The following sections describe each of the options in turn:

- *Using a CSV list of identifiers, below*
- *Creating a plate list, page 45.*

If one of the options is already selected, and you want to go back to using Auto-incrementing identifiers or manual identifiers:

Press



When you have selected an external source for plate identifiers, the icon in the **Plate List** box in the **Configuration** tab will show:



if you have selected to use an imported list or a list created in aCOLyte3



if you have chosen not to use an external source of plate identifiers.

### **Using a CSV list of identifiers**

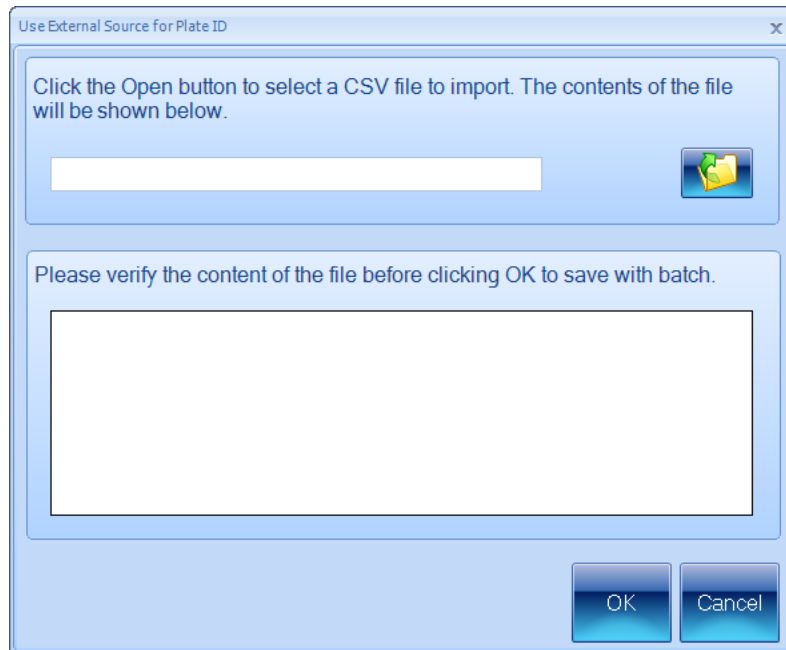
To use a CSV file containing a list of plate identifiers and dilutions (see the text following the instructions for the format to use for CSV files):

1. Press



## Creating a new batch

in the **Use External Source for Plate ID** dialog box (see *Using an external source for plate identifiers*, page 42) to display controls for opening the CSV file:

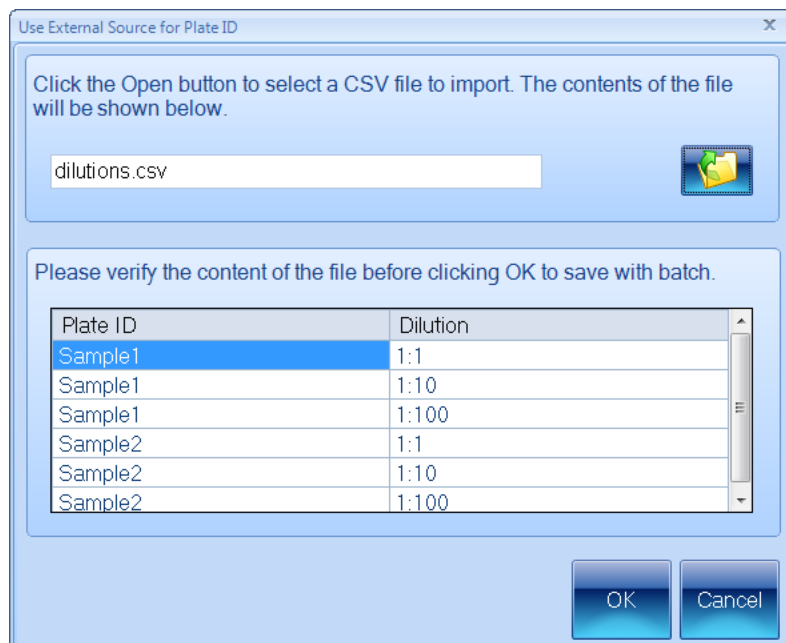


2. Press



to open a standard Windows **Open** dialog box.

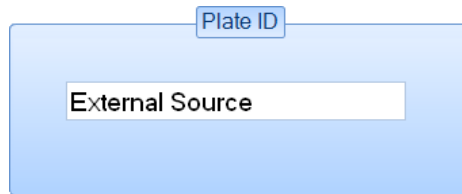
3. Use the **Open** dialog box to locate and select the required CSV file.
4. Press **Open** to open the selected file.
5. The contents of the file will be shown in the preview box:



Check the preview box to make sure the contents are correct (see the subsection following these instructions for the format to use for the CSV file).

6. If the contents are correct, press **OK** to close the **Use External Source for Plate ID** dialog box and load the plate list into the batch.

The **Plate ID** box on the **Measure tab** (see *The Measure tab*, page 47) will show that the plate identifiers have been taken from an external source:



---

**Note** When you are working in Measurement mode, you can use a barcode reader in conjunction with a CSV list to match each plate you are measuring to its identifier/dilution in the CSV list – see *Using a barcode reader for plate identifiers*, page 41.

---

### CSV file format

Each line of the file should consist of two strings separated by a comma:

*Identifier, Dilution*

where:

*Identifier* is the plate identifier

*Dilution* is text describing the dilution (if the string is not recognized or no dilution is given, 'No Dilution' is used).

The order of the lines in the file defines the order of the plates in the list.

For example:

```
Test1, No Dilution
```

```
Test2, 1:5
```

```
Test3, 1 in 1000
```

```
Test4, 1 in 10^6
```

---

**Note** aCOLyte3 does not *require* you to use unique identifiers/dilutions.

---

### Creating a plate list

To create a plate list containing a list of plate identifiers and dilutions:

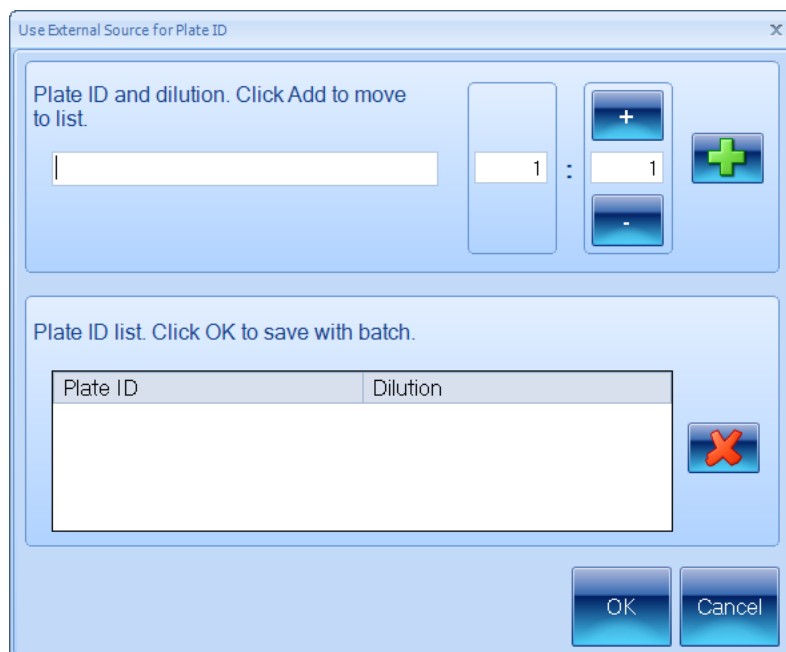
1. Press



## Creating a new batch

---

in the **Use External Source for Plate ID** dialog box (see *Using an external source for plate identifiers*, page 42) to display controls for creating a plate list:



2. Type the identifier for the first plate into the edit box at the top of the dialog box.
3. Use the dilution boxes to enter the dilution: you can type directly into the boxes or use the + and – buttons to increase or decrease the values.

---

**Note** The left-hand box is the sample proportion; the right-hand box is the total, so 1:10 means one part sample in ten parts total volume. This means that the right-hand number must not be smaller than the left-hand number.

---

4. Press



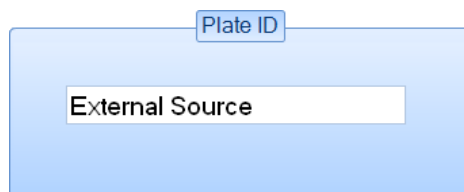
to add the identifier/dilution to the plate list.

5. Repeat Steps 2–4 for the remaining plates in the batch.
6. If you change your mind about one of the plates in the list, click on it in the list to select it and press



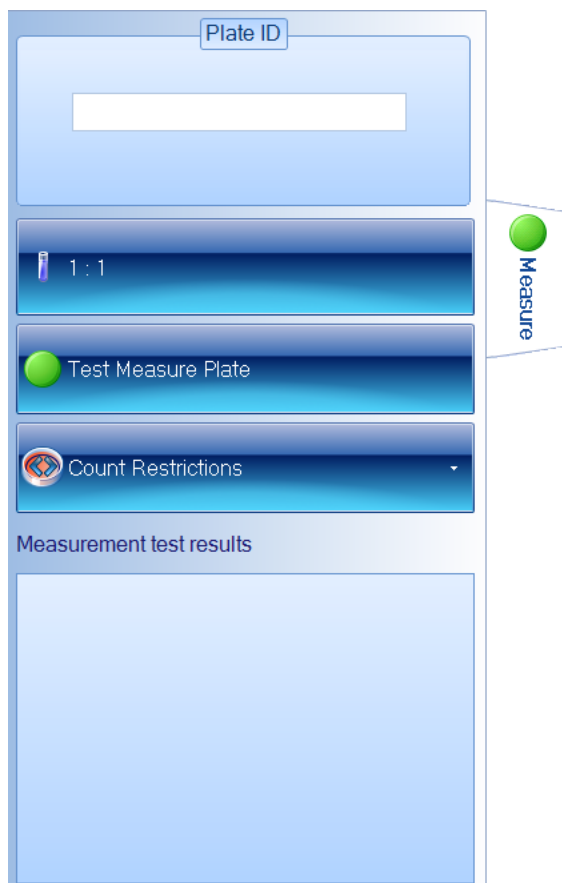
7. When you are satisfied that the list is complete and correct, press **OK** to close the **Use External Source for Plate ID** dialog box and load the plate list into the batch.

The **Plate ID** box on the **Measure** tab (see *The Measure tab*, on the facing page) will show that the plate identifiers have been taken from an external source:



**Note** When you are working in Measurement mode, you can use a barcode reader in conjunction with a custom plate list to match each plate you are measuring to its identifier/dilution in the plate list – see *Using a barcode reader for plate identifiers*, page 41.

## The Measure tab



**Notes** The picture shows the Measure tab for a Pour Plate batch: for Spiral batches, the dilution control has a different form (see *Spiral Plate batches*, page 50) and the **Count Restrictions** button is hidden.

To display the measure controls in the Batch Designer:

Press the **Measure** tab:



The **Measure** tab in the Batch Designer allows you to:

- enter text to act as a template for use with Auto-incrementing identifiers (see *Selecting auto-incrementing plate identifiers*, page 39)
- set the dilution ratio (see *Dilution*, on the next page)
- set count restrictions for Pour Plate batches – see *Setting count restrictions*, page 52

## Creating a new batch

---

- carry out a test count of the current plate (see *Test Measure Plate*, page 53) so that you can check the batch settings.

### Plate ID



The **Plate ID** control on the **Measure** tab in the Batch Designer allows you to specify a template for auto-incrementing plate identifiers – see *Selecting auto-incrementing plate identifiers*, page 39, for details.

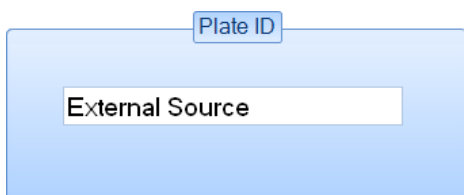
---

**Notes** If you choose to use auto-incrementing plate identifiers, you will not be able to accept the batch design until you have entered a template for the identifiers (see *Accepting the batch design*, page 56).

If you choose to use manually entered plate identifiers, you can enter a **Plate ID** in the Batch Designer, but it will be ignored when you accept the batch for measurement mode.

---

If you have chosen to use an imported plate list or a plate list created in aCOLyte3, the **Plate ID** control will show



### Dilution

---

**Notes** If you have chosen to use a CSV file or a custom plate list as the source of the plate identifiers for use in Measurement mode (see *Using an external source for plate identifiers*, page 42), you can include individual dilutions with each plate identifier – see the previous section, *Plate ID*. If no dilution is supplied in the lists, 'No dilution' is assumed.

---

To specify the dilution to use in calculating results:

Press the **Measure** tab



to display the dilution controls.

The dilution controls used for Pour Plate and Spiral Plate batches are different – for details, see:

- *Spiral Plate batches*, page 50
- *Pour Plate batches*, on the facing page.

---

### ***Pour Plate batches***

---

**Note** See *Spiral Plate batches*, on the next page, for how to set the dilutions for Spiral Plate batches.

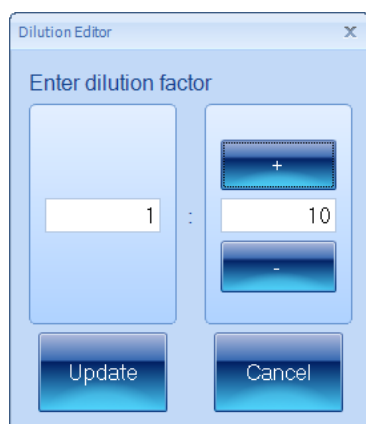
---

To set the dilution for a Pour Plate batch:

1. Press



(the button shows the current dilution) to open the dilution editor:



2. Use the dilution boxes to enter the dilution: you can type directly into the boxes or use the + and – buttons to increase or decrease the values.

---

**Note** The left-hand box is the sample proportion; the right-hand box is the total, so 1:10 means one part sample in ten parts total volume. This means that the right-hand number must not be smaller than the left-hand number.

---

3. Press **Update** to confirm the dilution you have set and return to the main aCOLyte3 window.

## Creating a new batch

---

### *Spiral Plate batches*

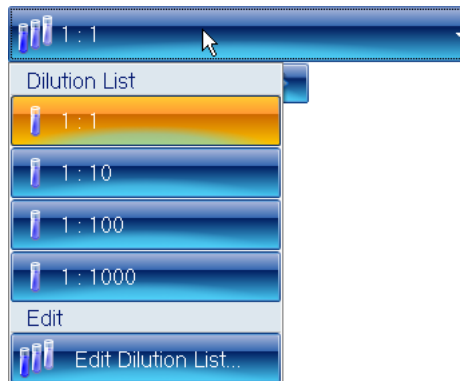
---

**Note** See *Pour Plate batches*, on the previous page, for how to set the dilutions for Pour Plate batches.

---

To set the dilutions for a Spiral Plate batch:

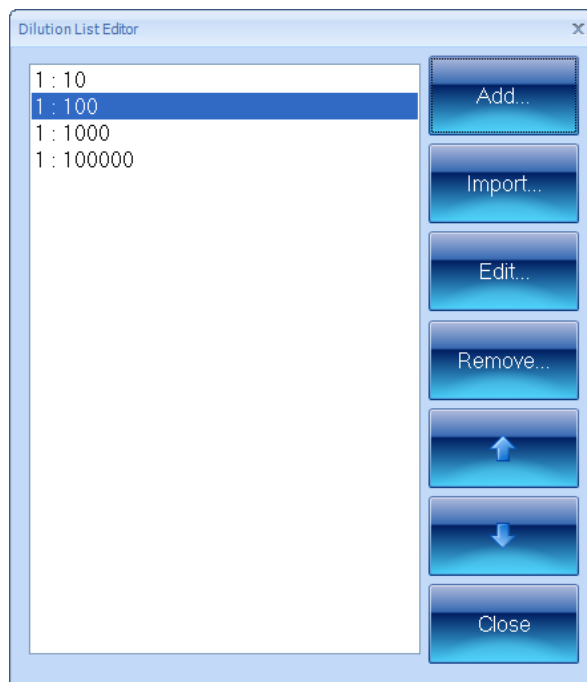
1. Press the dilutions button to display the dilutions menu (the button shows the current dilution):



**Note** The menu contains the dilutions in the current dilution list for the batch.

---

2. Select **Edit Dilution List** from the menu to open the **Dilution List Editor**:

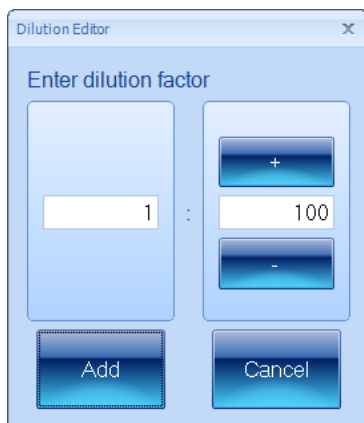


3. If required, to import a list of dilutions (see *Dilutions file format* after these instructions for details):
  - a. Press **Import** to open a standard Windows **Open** dialog box.
  - b. Use the **Open** dialog box to locate and select the required file.
  - c. Press **Open** to open the selected file.



The dilution list in the file will be added to the **Dilution List Editor**, replacing any existing entries.

4. To add a new dilution to the list:
  - a. Click on the dilution at the point you want to add the new dilution.
  - b. Press **Add** to open the **Dilution Editor**:



- c. Use the dilution boxes to enter the dilution: you can type directly into the boxes or use the + and – buttons to increase or decrease the values.

---

**Note** The left-hand box is the sample proportion; the right-hand box is the total, so 1:10 means one part sample in ten parts total volume. This means that the right-hand number must not be smaller than the left-hand number.

---

- d. Press **Add** to close the **Dilution Editor** and add the dilution to the list.
5. To change a dilution in the list:
  - a. Click on the dilution in the list to select it.
  - b. Press **Edit** to open the **Dilution Editor**.
  - c. Use the dilution boxes to edit the dilution: you can type directly into the boxes or use the + and – buttons to increase or decrease the values.
  - d. Press **Update** to close the **Dilution Editor** and confirm the change to the dilution.
6. To remove a dilution from the list:
  - a. Click on the dilution in the list to select it.
  - b. Press **Remove**.
 

A confirmation dialog box will be displayed.
  - c. Press **Yes** to remove the dilution.
7. To move a dilution up or down in the list in order to change the order of dilutions:
  - a. Click on the dilution in the list to select it.
  - b. Press the up or down arrow button.
8. When you have finished editing the dilutions list, press **Close** to return to the main aCOLyte3 window.

## Creating a new batch

---

**Note** When you carry out a test measurement for a Spiral Plate batch in the Batch Designer, the dilution selected in the dilutions menu affects the results, which show a count/ml value – see *Test Measure Plate*, on the facing page.

---

### Dilutions file format

Each line of the file should consist of a string representing the dilution (if the string is not recognized or no dilution is given, 'No Dilution' is used).

The order of the lines in the file defines the order of the dilutions in the list.

For example:

No Dilution

1:5

1 in 1000

1 in 10<sup>6</sup>

---

**Note** aCOLyte3 does not *require* you to use unique dilutions.

---

## Setting count restrictions

---

**Note** The **Count Restrictions** button is hidden for Spiral Plate batches.

---

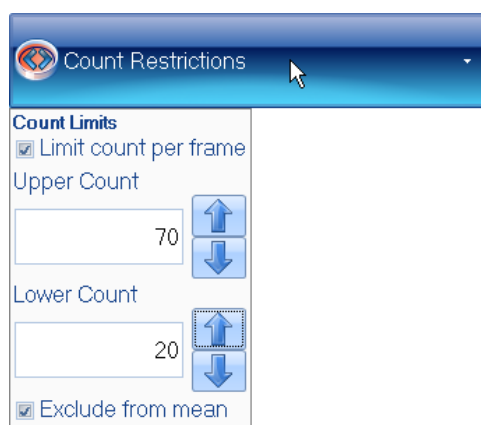
To place an upper limit on the count result:

1. Press the **Measure** tab



to display the measure controls, including the **Count Restrictions** button.

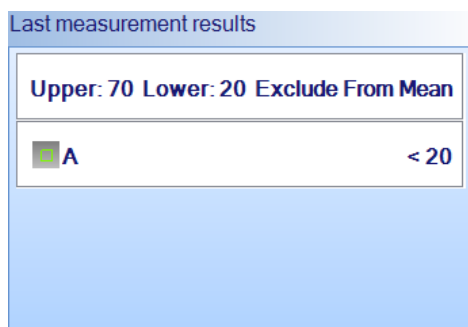
2. Press the **Count Restrictions** button to display the **Count Restrictions** controls:



3. Check **Limit count per frame** to enable count restrictions.
  4. Either type the required limits directly into the **Upper Count** and **Lower Count** edit boxes or press the arrow buttons to increase or decrease the values.
-

5. Check **Exclude from mean** if you want any results violating the count restrictions to be ignored when calculating the mean results; leave it unchecked to include all results in mean calculations.

When count restrictions have been set and you carry out a test measurement in the Batch Designer (see *Test Measure Plate*, below) or perform a measurement in Measurement mode, the **Measurement test results** or **Last measurement results** box, respectively, will show whether the limit has been breached, for example:



Unlike the **Last measurement results** box, which only shows that the limit has been breached, the Results table also records the actual count result.

## Test Measure Plate

To carry out a test measurement:

1. Press the **Measure** tab



to display the measure controls.

2. Press



**Note** If you are using an uncalibrated imported image, a warning dialog box will be displayed asking you to calibrate the image – see *Calibrating the image*, page 12, for details.

The results of the measurement will be shown in the **Measurement test results** panel.

If the button below the image is labeled **All Colony Markers**, the detected colonies will also be marked on the image using the selected colors and markers (if the button is labeled **No Colony Markers**, press the button and select **All Colony Markers** to display the detected colonies on the image).

For more detailed information about the precise areas identified as colonies, you can also select:

- **All Colony Outlines:** the boundary of each detected colony is marked in the color of the symbol representing the type of colony

## Creating a new batch

---

- **All Colony Regions:** the area of each detected colony is marked in the color of the symbol representing the type of colony.

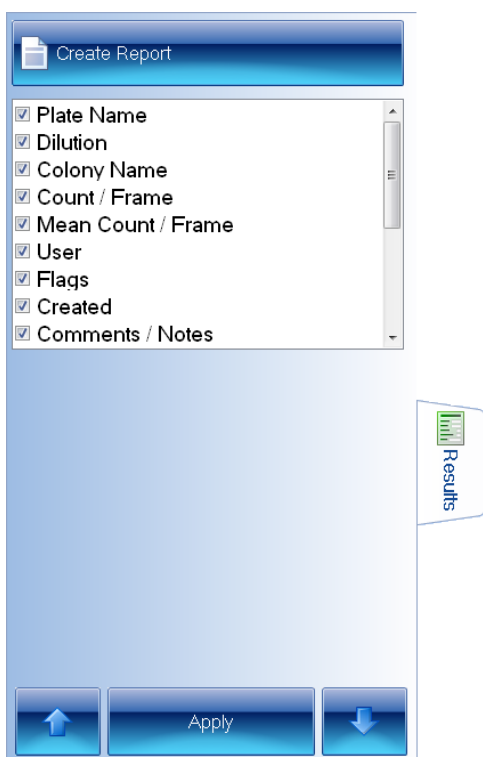
If required, you can zoom the image to check the colony detection – see *Zooming the image*, page 10.

3. Check the image to make sure that the colonies have been correctly detected and distinguished from the background and any debris on the plate.
4. If there are any problems with the colony detection, you will need to go back to change the batch settings using one of the procedures described earlier in *Total Plate Count*, page 30.

If you are happy with the measurement results, you can proceed to accept the batch for Measurement mode – see *Accepting the batch design*, page 56.

## The Results tab

The **Results** tab allows you to specify what results should be included in the Results table and the order of the columns:



---

**Notes** The picture shows the **Results** tab in Measurement mode; the **Apply** button does not appear in the Batch Designer.

The results included in the list depend on the type of plate used in the batch.

---

To display the results controls:

Press the **Results** tab:



You can set a default selection of results in the Batch Designer, but you can also make a different selection at any time in Measurement mode as required.

Results with a selected check box will be included in the Results table; unselected results will not be included. The order of the results in the list is the order they will appear in the Results table.

To change whether a result will be included in the Results table:

1. Click on the result to select it.
2. Click on the result again to select/deselect the check box.
3. To carry out the change when working in Measurement mode, press:



To move a result up or down the list to change the order of results in the Results table:

1. Click on the result to select it.
2. To move the result up the list, press



To move the result down the list, press



3. To carry out the change when working in Measurement mode, press:



### Accepting the batch design

Once you have accepted a batch for use in Measurement mode (see *Accepting the batch design*, above), you will not be able to change whether aCOLyte3 detects light or dark colonies. However, you will be able to carry out a test measurement and change the settings for the **Reject Small Particles** and **Sensitivity** sliders and the colony splitter before recording the result (see *Test measurement and adjusting settings*, page 72). You can also edit a result by adjusting these settings after you have recorded it – see *Rejecting small particles in a result*, page 84, *Changing the Sensitivity setting for a result*, page 85, and *Splitting colonies in a result*, page 85, for details. You can also change the colony markers and colors in Measurement mode (see *The Classification panel*, page 33).

To confirm that you have completed the setup procedure and want to proceed to taking measurements:

Press



---

**Notes** The button will be disabled until you have carried out all steps required for setting up the batch. However, even if the button is enabled, you will be prevented from accepting the batch if you have not given the batch a name (see *Giving the batch a name*, page 25) or, if you are using auto-incrementing plate identifiers (see *Selecting auto-incrementing plate identifiers*, page 39), you have not set a template plate id (see *Plate ID*, page 48).

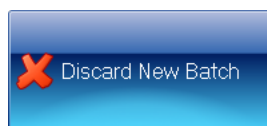
The **Accept New Batch** button is enabled to show that you *can* proceed to taking measurements if you wish – you can still make further changes to the setup if required before pressing the **Accept New Batch** button.

---

The next chapter in the Manual, *Using Measurement Mode to count colonies*, shows you how to use aCOLyte3 to carry out colony counts; the chapter following that, *Working with results*, page 77, shows you how to view and edit the results, and then compile a report.

If you change your mind about creating the new batch:

Press



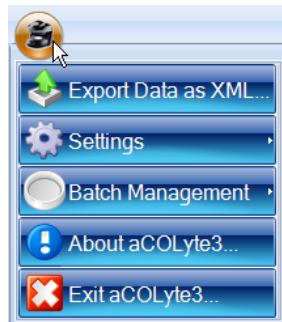
to abort the new batch – you will be asked to confirm that you want to do this.

## Managing batches

### Exporting batch details

To export the details of a batch to an XML file:

1. Press the aCOLyte3 button at the top left-hand corner of the window to display the aCOLyte3 menu:



2. Press:



to display the **Export Data** dialog box – this is a standard Windows **Save As** dialog box.

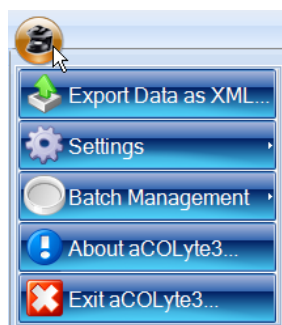
3. Use the **Export Data** dialog box to select a location and enter a filename for the exported batch details.
4. Press **Save** to save the batch details file.

If required, you can move the XML file to another PC and use it as the basis for a new batch on that PC – see *Creating a new batch based on an existing batch or an exported batch file*, page 21.

### Deleting batches

To delete a batch from the database:

1. If the batch you want to delete is currently open, close it – see *Closing batches*, page 75.
2. Press the aCOLyte3 button at the top left-hand corner of the window to display the aCOLyte3 menu:



## Creating a new batch

---

3. Press:

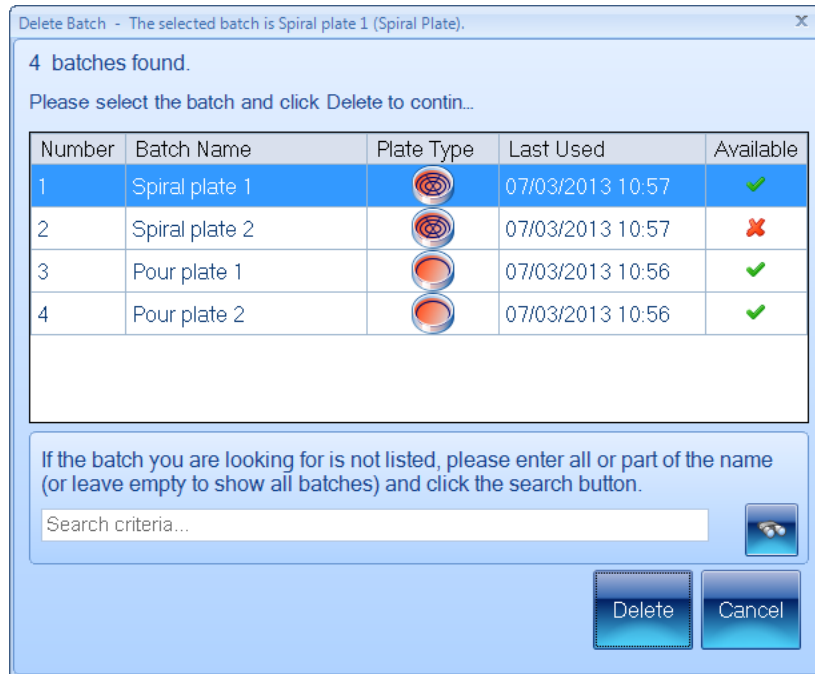


to display the **Batch Management** submenu.

4. Press:



to display the **Delete Batch** dialog box listing **The 10 most recent batches**:



5. If the required batch is not in the list of most recent batches:
- Type the name, or a part of the name, of the required batch into the **Search criteria** box.
  - Press

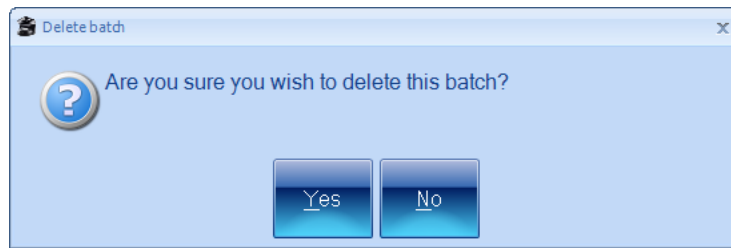


Only batches with names that contain the text you entered will be listed.

- Select the required batch in the list – you will only be able to delete batches if they are ticked in the **Available** column; open batches will not be available.
- Press **Delete** to delete the batch.

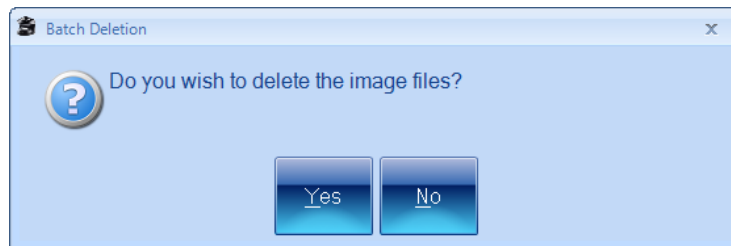


You will be asked to confirm that you want to delete the batch:



8. Press **Yes** to delete the batch.

You will be asked if you want to delete the image files:



9. Press **Yes** to delete the image files saved with the batch.

---

**Note** Although the batch can no longer be viewed in aCOLyte3, it is not actually removed from the aCOLyte3 database and can be viewed there if required for audit purposes.

---



# Using Measurement Mode to count colonies

---

Once you have created and accepted a new batch (see *Creating a new batch*, page 17, and *Accepting the batch design*, page 56), the general procedure for counting colonies is:

1. Open or select the batch – see the next section, *Opening and selecting batches*.
2. Load the first plate into aCOLyte3 – see *Loading plates into the instrument*, page 8.
3. Capture an image – see *Capturing an image*, page 9.
4. Adjust the measurement frame(s) if required – see *Adjusting frame settings in Measurement mode*, page 63.
5. Enter a plate identifier if it has not been entered automatically, or change an automatically entered identifier and dilution if required – see *Entering plate identifiers and dilutions*, page 63.
6. Carry out the measurement – see *Counting colonies*, page 68.
7. Repeat Steps 2–6 for all the other plates in the batch.
8. Check the results and edit them if necessary, then create a report for the batch – see the following chapter, *Working with results*, page 77.
9. Close the batch – see *Closing batches*, page 75.

Steps 2 and 3 are also required when creating and setting up the batch – the cross references take you back to earlier sections in the Manual. Step 8 is described in the next chapter, *Working with results*, page 77.

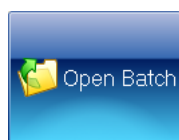
Instructions for the remaining steps are given in this chapter (though much of Step 4 is common to creating and setting up the batch and is covered by references to the *Creating a new batch*, chapter).

## Opening and selecting batches

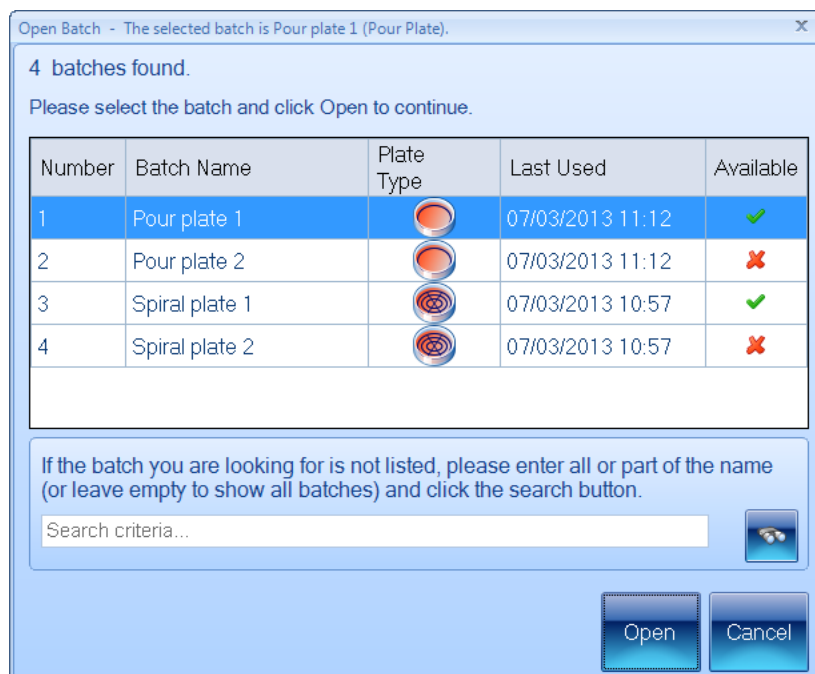
If you have just created and accepted a new batch (see *Creating a new batch*, page 17, and *Accepting the batch design*, page 56), it will be automatically open and selected in Measurement mode. However, you can also open a previously created batch if there is no batch open currently, or if you want to open another batch in addition to any currently open batches (you can have several batches open at the same time – see later in this section for how to select which open batch to work with).

To open a batch in Measurement mode:

1. Press:



to display the **Open Batch** dialog box listing **The 10 most recent batches**:



Open batches are marked with a ✗ in the **Available** column.

2. If the required batch is not in the list of most recent batches:
  - a. Type the name, or a part of the name, of the required batch into the **Search criteria** box.
  - b. Press



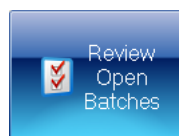
Only batches with names that contain the text you entered will be listed.

3. Select the required batch in the list.
4. Press **Open** to open the batch.

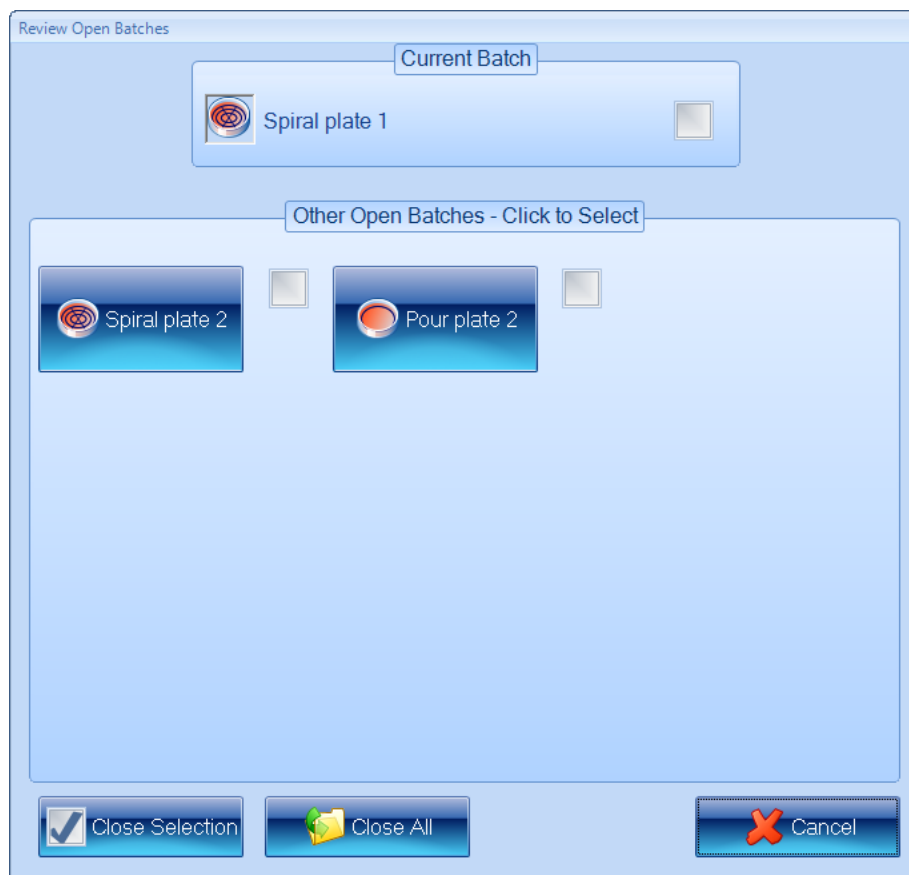
If there is more than one batch currently open in aCOLyte3, you can select which one to work on at any time (unless you are in the process of creating a new batch).

To select an open batch:

1. Press:



to display the **Review Open Batches** dialog box:



The currently selected batch is listed with a check box in the **Current Batch** box, and there are buttons and check boxes for the **Other Open Batches** in the box below it.

2. Press the button for the required batch in the **Other Open Batches** box.

The **Review Open Batches** dialog box will close and the selected batch shown in Measurement mode.

## Adjusting frame settings in Measurement mode

If required, you can adjust the frames settings in Measurement mode by changing: the frame type, shape, size, position and orientation for an individual plate before you make a plate measurement. You use essentially the same techniques as when you are setting up a batch – see the following for details:

- Frame shape – see *Choosing the type of frame to use*, page 26
- Frame size, position and orientation – see *Adjusting the position and size of frames*, page 28.

## Entering plate identifiers and dilutions

Each plate in the batch must be given a plate identifier, but the procedure required depends on the options selected when the batch was created.

## Using Measurement Mode to count colonies

---

**Note** If several plates are given the same identifier (for example, with different dilutions of the same sample), they will be grouped together under the same item in the Results table – see *Working with results*, page 77, for details.

---

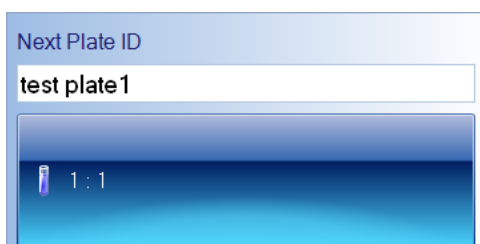
For details, see:

- the next section, *Auto-incrementing plate identifiers*
- *Manually entered plate identifiers*, below
- *External source – CSV file or Custom plate list*, page 66
- *Using a barcode reader for identifiers and dilutions*, page 68.

### Auto-incrementing plate identifiers

See *Selecting auto-incrementing plate identifiers*, page 39, for details of how to set up a batch to use auto-incrementing plate identifiers, including the rules used to generate the new identifier.

When a batch with auto-incrementing plate identifiers is first opened, the template identifier set when the batch was created (see *Selecting auto-incrementing plate identifiers*, page 39) will be set automatically in the **Next Plate Id** box as the first identifier for the batch. For example:



After each measurement, a new plate identifier will be set ready for the next plate by auto-incrementing the previous identifier.

If required, you can edit the identifier in the **Next Plate Id** box. That identifier will then be used for the next measurement, and will be auto-incremented for subsequent measurements according to the normal auto-incrementing rules.

### Manually entered plate identifiers

To enter a manual plate identifier:

Type the identifier into the **Next Plate ID** box:



You can also use a barcode reader to enter a manual plate identifier (and dilution if the barcode reader is programmed to read dilutions).

## Changing the dilution set for a plate

If required, you can change the dilution set for the current plate. The procedure required depends on the type of batch.

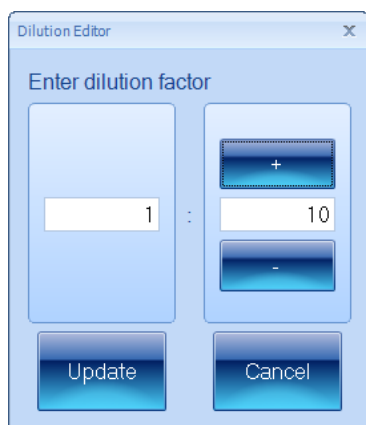
### Pour Plate batches

To change the dilution for the current plate:

1. Press the dilution button, for example:



to open the **Dilution Editor**:



2. Use the dilution boxes to enter the dilution: you can type directly into the boxes or use the + and – buttons to increase or decrease the values.

**Note** The left-hand box is the sample proportion; the right-hand box is the total, so 1:10 means one part sample in ten parts total volume. This means that the right-hand number must not be smaller than the left-hand number.

3. Press **Update** to confirm the dilution you have set and return to the main aCOLyte3 window.

### Spiral Plate batches

If you have already created a list of dilutions for the batch (see later in this section for how to create a new dilutions list), to select a different dilution from the list:

Press one of the arrow buttons below the dilution button to select the next or previous dilution in the series, for example:

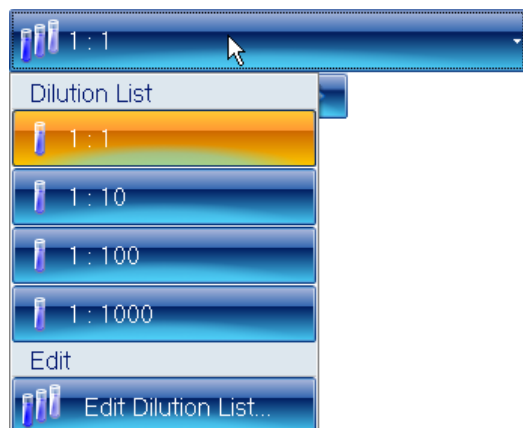


## Using Measurement Mode to count colonies

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Or:

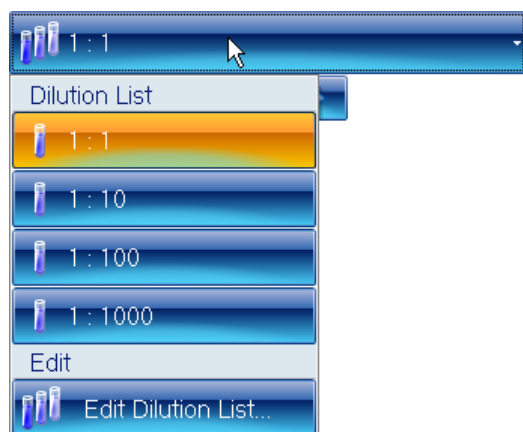
1. Press the dilution button, for example:



2. Select the required dilution from the **Dilution List**.

To edit the dilution list and choose a dilution from the new list:

1. Press the dilution button, for example:



2. Press **Edit Dilution List** to display the **Dilution List Editor**.
3. Use the **Dilution List Editor** to edit the dilution list – see *Spiral Plate batches*, page 50, for how to use this dialog box.
4. Press the dilution button again and select the required dilution from the **Dilution List**.

---

**Note** The new dilution list will be used for future plates as well as the current one.

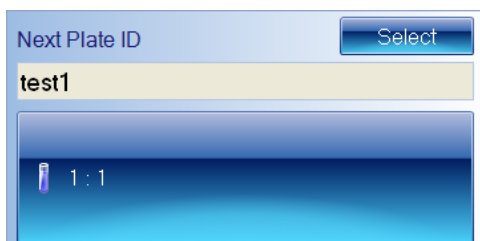
---

### External source – CSV file or Custom plate list

When the batch is set to use an external source of plate identifiers (see *Using an external source for plate identifiers*, page 42), aCOlyte3 creates a list of plate identifiers and, if relevant, dilutions from the external source.



When you first open the batch, the **Next Plate ID** box shows the first plate identifier on the list and, where relevant, the dilution control shows the dilution, for example:



**Note** The **Next Plate ID** text box is disabled for a batch using an external source for its plate identifiers – you cannot edit the identifiers.

After you have performed the measurement, the identifier (and dilution where relevant) for that measurement will be removed from the plate list and the next identifier (and dilution) will be shown. You can then repeat this procedure until the list is exhausted. When the list is exhausted, no more measurements can be made and an error message will be displayed if you try to perform a count.

If the plates are not arranged in the same order as the plate list, you can choose another identifier/dilution from the list – see the following subsection.

### Selecting a different plate from the plate list

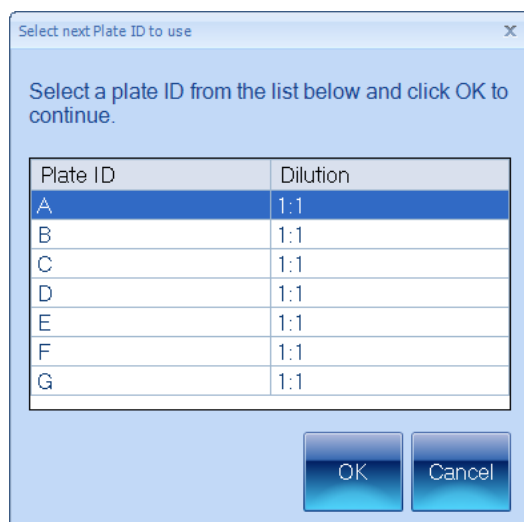
If the batch has been set up for use with a barcode reader (see *Using a barcode reader for plate identifiers*, page 41), you can use the barcode reader to read the plate identifier (and dilution where relevant and if the barcode reader is programmed to read dilutions) from the plate – aCOLyte3 will give an error if the identifier/dilution is not on its plate list for the batch.

Alternatively, if you need to choose a different plate identifier/dilution from the plate list:

1. Press



to display the **Select next Plate ID to use** dialog box:



## Using Measurement Mode to count colonies

---

2. Click in the row containing the required plate identifier/dilution to select it.
3. Press **OK** to close the dialog box – the selected identifier will appear in the **Next Plate Id** box and the dilution will appear on the dilution button.

## Using a barcode reader for identifiers and dilutions

You can use a barcode reader to enter a manual plate identifier (and dilution if the barcode reader is programmed to read dilutions) or pick a plate identifier and dilution from the plate list created from an external source (see *Using an external source for plate identifiers*, page 42).

## Counting colonies

This section of the Manual describes how to count the colonies on a plate in Measurement mode. In most cases, you should be able to carry out an automatic colony count by simply pressing a button – for details, see *Counting the colonies on a plate automatically*, on the facing page.

However, some plates or batches may require individual attention. In particular, you can:

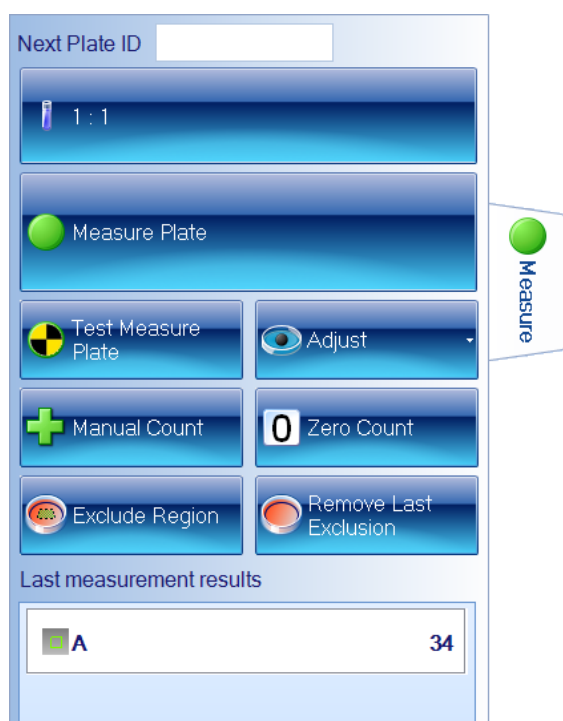
- Create exclude regions to avoid areas of the plate that might cause problems – see *Exclude regions*, on the facing page
- Carry out a test measurement and adjust the colony detection settings – see *Test measurement and adjusting settings*, page 72
- Carry out a manual colony count – see *Manual colony counting*, page 74
- Enter a zero result for a blank plate – see *Entering a zero count*, page 75.

---

**Note** Some of the above procedures can only be applied to Pour Plate batches – all exceptions are noted in the instructions.

---

All the above colony counting procedures use the **Measure** tab:



**Note** The picture shows the **Measure** tab in Measurement Mode for a Pour Plate batch – the **Manual Count**, **Exclude Region** and **Remove Last Exclusion** buttons do not appear for Spiral Plate batches. The **Adjust** button is only enabled after you have pressed **Test Measure Plate** and the **Remove Last Exclusion** is only enabled if you have placed an exclusion region on the image.

To display the **Measure** tab in Measurement mode:

Press:



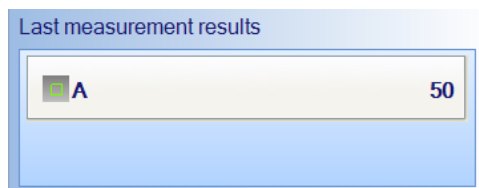
### Counting the colonies on a plate automatically

To count the colonies on a plate automatically:

Press



The results will be added to the Results table (see *Working with results*, page 77, for details) and shown in the **Last Measurement Results** panel:



For a Spiral Plate batch, the **Last Measurement Results** will also show the concentration:



**See also:**

*Counting colonies*, on the previous page, for a summary of how to modify the way automatic colony counts are performed and how to carry out colony counts using other methods.

### Exclude regions

**Note** You cannot place exclude regions on Spiral Plate batches.

On occasions, you may want to avoid counting colonies on part of a plate because of some problem with it. One way of doing this is to create one or more exclude regions, which will be

## Using Measurement Mode to count colonies

---

ignored when aCOLyte3 counts the colonies – the area of the exclude regions will be deducted from the area of the frame to ensure that results such as count/ml are correct.

---

**Note** An alternative for Pour Plate batches might be to choose a Partial Plate frame and/or to rotate the frame so that the problem area is not counted.

---

To define an exclude region on an image:

1. Press



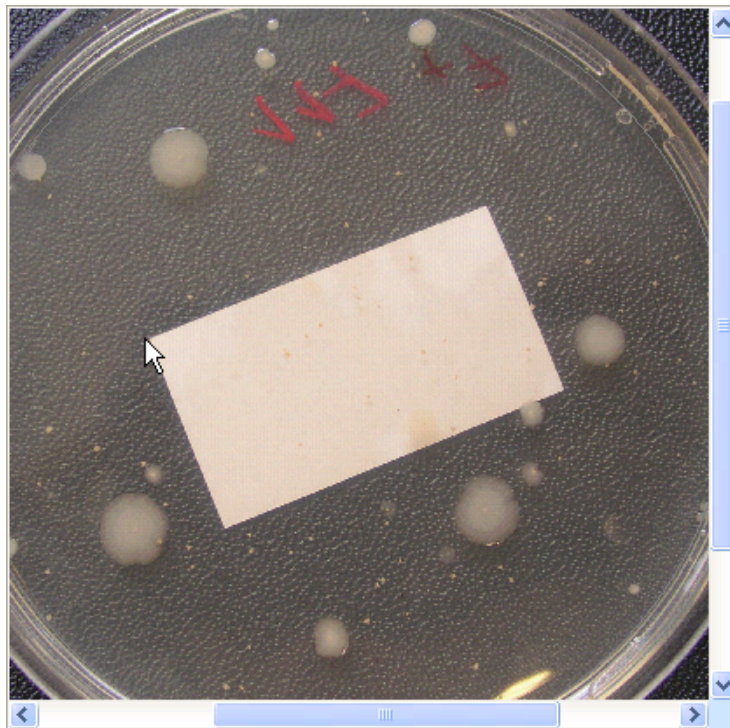
to select it:



**Note** You may find it helpful to zoom the image before placing the exclude region on the image – see *Zooming the image*, page 10.

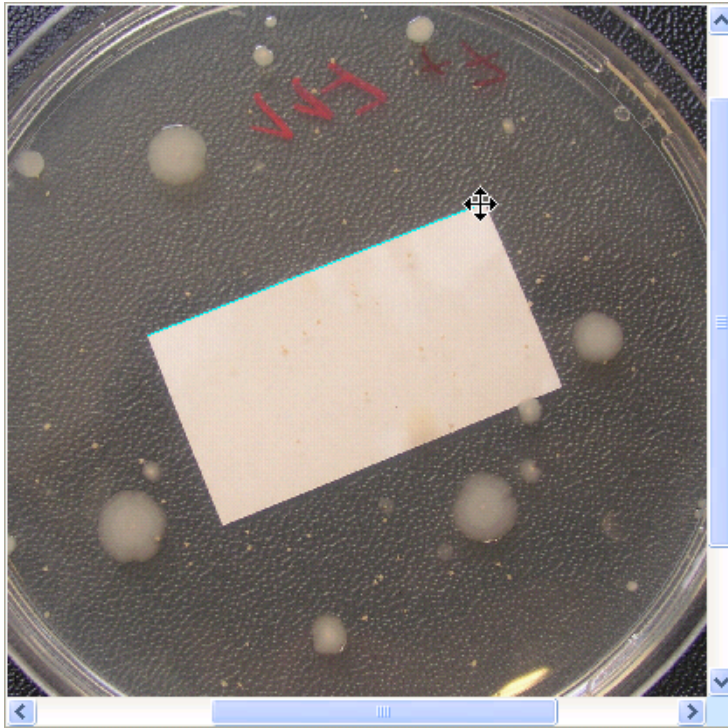
---

2. Move the pointer to the position you want to place the first vertex of the exclude region:

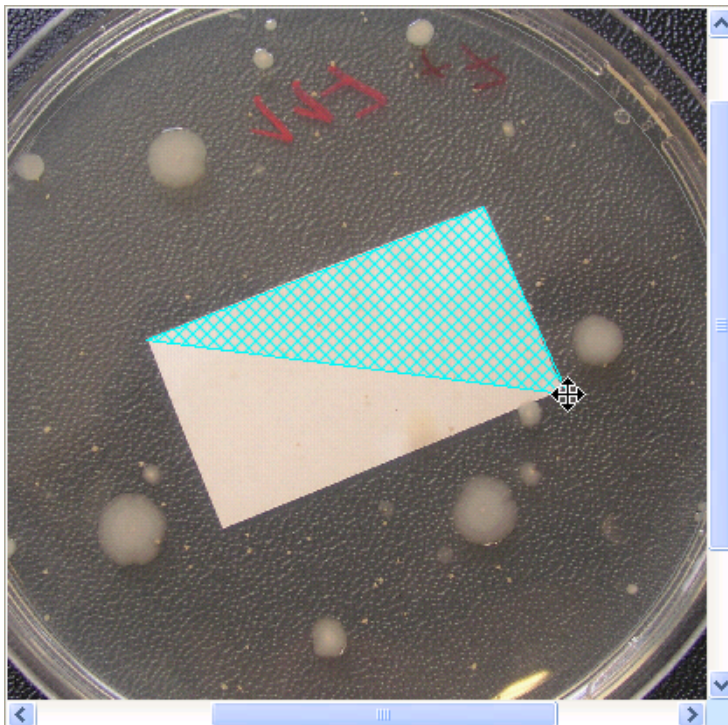


3. Click to place the first corner of the exclude region.

4. Move the pointer to the position you want to place the next vertex of the exclude region – a line will be dragged out from the first vertex:



5. Click to add the new vertex for the exclude region.
6. Repeat Steps 4 and 5, as required, to place any further vertices until you have placed the penultimate one (the included area is shown as you drag the mouse):



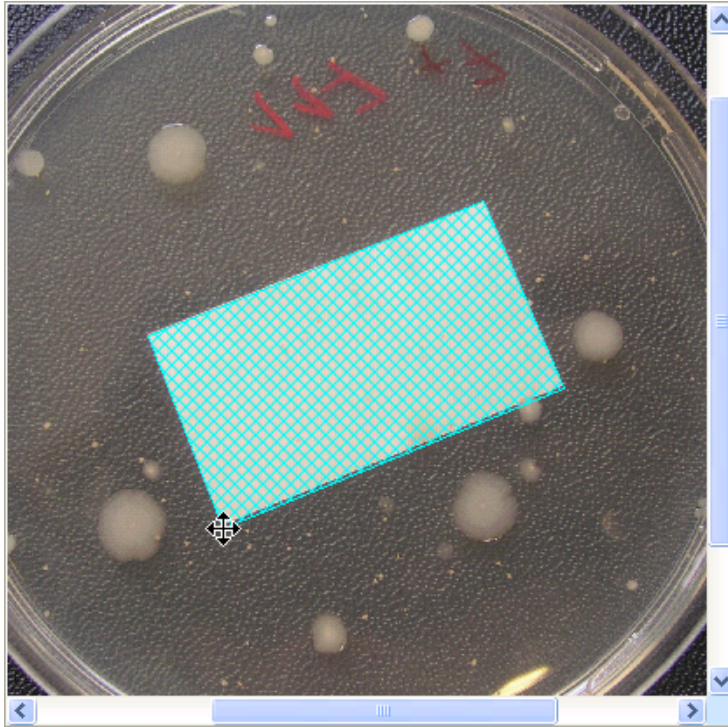
7. Move the pointer to the final vertex of the exclude region.



## Using Measurement Mode to count colonies

---

8. Double-click to add the vertex and complete the region.



To remove the last exclude region you added to the image:

Press



You can repeat these procedures to add and remove as many exclude regions as required.

### Test measurement and adjusting settings

If required, you can carry out a test measurement to check the colony detection for an individual plate without recording the result, and, if required, you will be able to modify the batch colony detection settings if there are any problems.

To carry out a test colony count and adjust the detection settings:

1. Press



The result of the test measurement will appear in the **Last Measurement Results** panel and the **Adjust** button will be enabled:

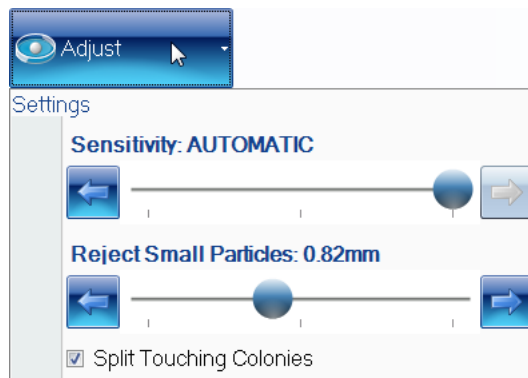


The result of the test measurement will **not** be added to the Results table (see *The Results table*, page 77).

If the button below the image is labeled **All Colony Markers**, the detected colonies will also be marked on the image using the colors and markers selected for each colony type (if the button is labeled **No Colony Markers**, press the button and select **All Colony Markers** to display the detected colonies on the image).

2. Check the image to make sure that the colonies have been correctly detected.
3. If you are satisfied with the colony detection, go straight to Step 4.

Otherwise, if there are problems with the colony detection, press **Adjust** to display the **Settings** menu:



The controls are used in exactly the same way as when you are setting up a batch – see *Total Plate Count*, page 30, for details.

**Note** If required, you will also be able to change the **Sensitivity**, **Reject Small Particles** and **Split Touching Colonies** settings after you have performed and recorded the count – see *Rejecting small particles in a result*, page 84, *Changing the Sensitivity setting for a result*, page 85, and *Splitting colonies in a result*, page 85, for details.

## Using Measurement Mode to count colonies

---

4. To carry out the measurement and record the results in the Results table, press



---

**Note** If you make any changes to the settings using the procedures described in this section, the new settings will be applied to the batch until you close the batch. If you wish to revert to the settings made when the batch was created, either make a note of the values and re-enter them using the above procedure, or close the batch (see *Closing batches*, on the facing page) and reopen it again (see *Opening and selecting batches*, page 61).

---

## Manual colony counting

---

**Note** You cannot carry out a manual colony count for a Spiral Plate batch.

---

To carry out a manual colony count:

1. Press



The button will become selected showing that a manual count is in progress:



2. Click on the image to count each of the colonies in turn – as you click, the total in the **Last measurement results** panel will be incremented and the image will be marked to show that the colony has been counted.
3. When the count is complete, press



to record the result in the results table.

---

**Notes** Once you have begun a manual count, the only way to cancel it is to log out of or close aCOLyte3.

You can only add to the total by clicking on the image; you cannot decrease the count.

---

### See also

*Manually adding and removing colonies*, page 83, for how to edit a result in the results table by adding or removing colonies **after** the count has been made and recorded.



### Entering a zero count

To enter a zero result for a plate:

Press



A new result will be added to the Results table with a 0 count.

---

**Note** The image will not be saved with the result, even if **Save images to file** is selected in the **Settings** dialog box – see *System settings*, page 92.

---

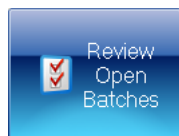
#### See also

*Zeroing the count*, page 87, for how to zero a result in the results table **after** the count has been made and recorded.

### Closing batches

To close one or more open batches:

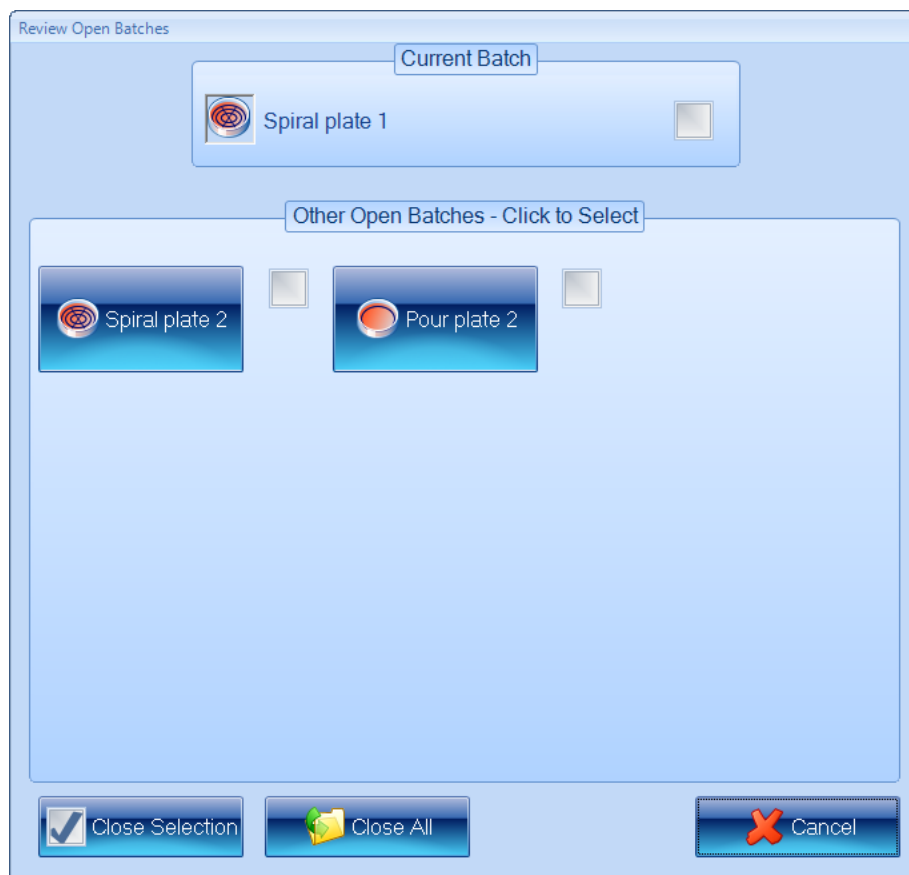
1. Press:



## Using Measurement Mode to count colonies

---

to display the **Review Open Batches** dialog box:



The currently selected batch is listed with a check box in the **Current Batch** box, and there are buttons and check boxes for the **Other Open Batches** in the box below it.

2. Either, to close a selection of batches:
  - a. Click in the check boxes for the batches you want to close to select them.
  - b. Press



Or, to close all the open batches:

Press



The **Review Open Batches** dialog box will close and either the selected batches or all the batches will be closed.

# Working with results

This chapter shows you how to view and process the results produced by aCOLyte3, and then compile a report based on them (see *Using Measurement Mode to count colonies*, page 61, for how to carry out the measurements). This chapter has three main subsections:

- *The Results table*, below
- *Editing plate measurements*, on the next page
- *Compiling reports*, page 88.

## The Results table

When aCOLyte3 has counted the colonies on a plate, it adds the results to the top of the Results table at the bottom of the window, for example:

Plate Name	Dilution	Colony Name	Count / Plate	Count / ml
Sample C	1 : 1	A	42	42
Sample B	1 : 1	A	100	100
Sample A				
Mean				
Colony Name		Mean Count / ml		
A		300		
Plates				
Plate Name	Dilution	Colony Name	Count / Plate	Count / ml
Sample A	1 : 10	A	28	280
Sample A	1 : 3	A	100	300
Sample A	1 : 1	A	246	246

**Note** The default choice of which columns are included and their order in the Results table is made when the batch is set up in the Batch Designer, but you can change this at any time in Measurement mode – see *The Results tab*, page 54 (in Measurement mode, you will need to press the **Apply** button to carry out any changes you make in the **Results** tab).



In the example, there are results for three Plate Names: **Sample A**, **Sample B** and **Sample C** – there is a single result each for **Sample B** and **Sample C**, but the Plate Name **Sample A** has been used for three plates with different dilutions, so it contains three separate **Plates** results together with the **Mean** result.

## Working with results

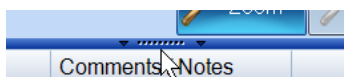
If there is more than one result for a Plate Name, you can choose how much information to show by opening or closing the **Plate Name**, the **Mean** result or the **Plates** details within it, for example:

Plate Name	Dilution	Colony Name	Count / Plate	Count / ml
Sample C	1 : 1	A	42	42
Sample B	1 : 1	A	100	100
Sample A				
Mean				
Colony Name		Mean Count / ml		
A		300		
Plates				

To open or close a plate name or some of the details within it:

Press the  or  button next to the item you want to open or close.

If required, you can resize the Results table by dragging the handle at the top of the table:





You can also resize the table columns by dragging the dividing line between the headings:



You can select a row in the Results table by clicking in it, and move to the next or previous line by pressing the down or up arrow key on the keyboard.

To select the next or previous **Plate Name** in the Results table (remember there may be several plates with the same **Plate Name**):

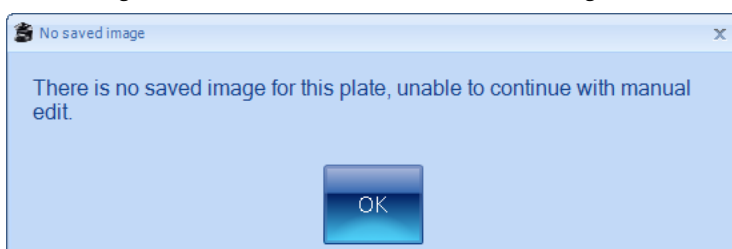
Press  or  on the right-hand edge of the Results table.

## Editing plate measurements

aCOLyte3 allows you to edit measurements in a variety of ways using the **Edit Result** tab.

**Notes** You can only edit results if the image was saved with the result – see *System settings*, page 92, for how to choose to save images with results.

If the image was not saved with the result, a dialog box will be displayed:



Press **OK** to continue.

To edit a measurement:

1. Select the measurement you want to edit by clicking in the plate's result in the Results table – see *The Results table*, page 77.
2. Press the **Edit Result** tab



to display the **Editing Plate** panel at the bottom of the aCOLyte3 window:

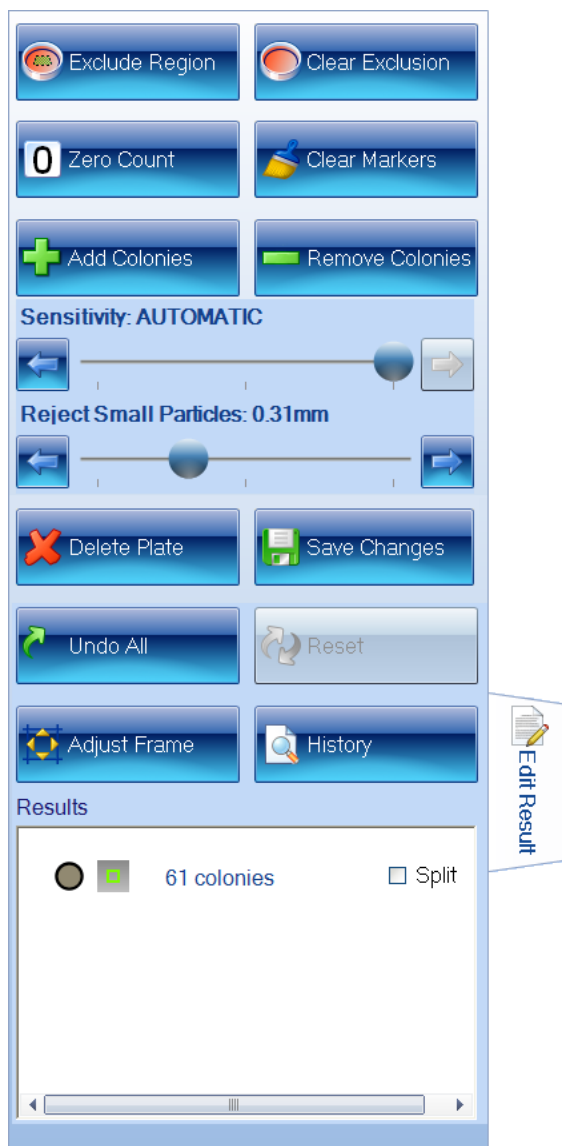
A screenshot of the "Editing Plate" panel in the aCOLyte3 software. It features a light blue background. On the left, the text "Editing Plate:" is followed by a white text box containing "plate 5". To the right of this is a blue button with a white upward-pointing arrow and the text "Dilution 1 : 3". Further right is a white text box with a blue border and a small "Comments" label above it.

and the **Edit Result** controls in the **Edit Result** tab.

## Working with results

---

The controls included on the **Edit Result** tab depend on the type of batch. For example, for a Pour Plate batch, you get the following controls:



---

**Notes** The **Exclude Region**, **Clear Exclusion** and **Adjust Frame** controls are hidden for Spiral Plate batches.

---

The controls on the **Edit Result** tab and the **Editing Plate** panel allow you to carry out a wide range of editing operations, which are described in the following sections:

- *Editing the plate identifier, comments and dilution, on the facing page*
- *Saving changes to results, page 82*
- *Undoing and resetting changes, page 82*
- *Deleting results, page 82*
- *Viewing the audit history of changes, page 83.*
- *Manually adding and removing colonies, page 83*

- *Adjusting the frame for a result*, page 84
- *Rejecting small particles in a result*, page 84
- *Changing the Sensitivity setting for a result*, page 85
- *Splitting colonies in a result*, page 85
- *Adding exclude regions to a result*, page 86
- *Zeroing the count*, page 87.

## Editing the plate identifier, comments and dilution

To add or edit a comment and/or edit the plate identifier and/or, where relevant, edit the dilution for a result:

1. Select the **Edit Result** tab for the plate result – see *Editing plate measurements*, page 78.

The **Editing Plate** panel will be displayed at the bottom of the aCOLyte3 window:



2. Edit the plate identifier in the left-hand edit box as required.
3. Add to/edit the **Comments** in the right-hand edit box as required.
4. To edit the dilution, where relevant:
  - a. Press the dilution button, for example:



to open the **Dilution Editor**:



- b. Use the dilution boxes to enter the dilution: you can type directly into the boxes or use the + and – buttons to increase or decrease the values.

---

**Notes** The left-hand box is the sample proportion; the right-hand box is the total, so 1:10 means one part sample in ten parts total volume. This means that the right-hand number must not be smaller than the left-hand number.

---

- c. Press **OK** to confirm the dilution you have set and return to the **Editing Plate** panel.
5. Save the change to the result (see *Saving changes to results*, on the next page).

## Working with results

---

The identifier, comment and/or dilution will be updated for the result and if you have changed the dilution, values depending on the dilution will also be updated. An **E** flag will appear in the **Flags** column showing the result has been edited.

### Saving changes to results

To save any changes you have made to a result on the **Edit Result** tab and/or the **Editing Plate** panel (see *Editing plate measurements*, page 78):

Press



---

**Note** Any changes you have made to a result will be lost if you move away from the **Edit Result** tab before saving them.

---

If required, you can view an audit history listing the changes you have made to the result – see *Viewing the audit history of changes*, on the facing page.

### Undoing and resetting changes

To undo all the changes made to a result on the **Edit Result** tab and the **Editing Plate** panel (see *Editing plate measurements*, page 78) since the result was last saved (see *Saving changes to results*, above):

Press



To reset the result to the original measurement value before any changes were made to it and saved using the **Edit Result** tab:

Press



---

**Note** Changes made to the plate identifier, dilution or comment in the **Editing Plate** panel will not be reset.

---

### Deleting results

To delete the result(s) for a plate:

1. Select the **Edit Result** tab for the plate result – see *Editing plate measurements*, page 78.
2. Press



3. Save the change to the result (see *Saving changes to results*, above).

The result will be deleted from the Results table.



**Note** Although the result is deleted from the Results table and can no longer be viewed in aCOLyte3, it is not removed from the aCOLyte3 database and can be viewed there if required for audit purposes.

### Viewing the audit history of changes

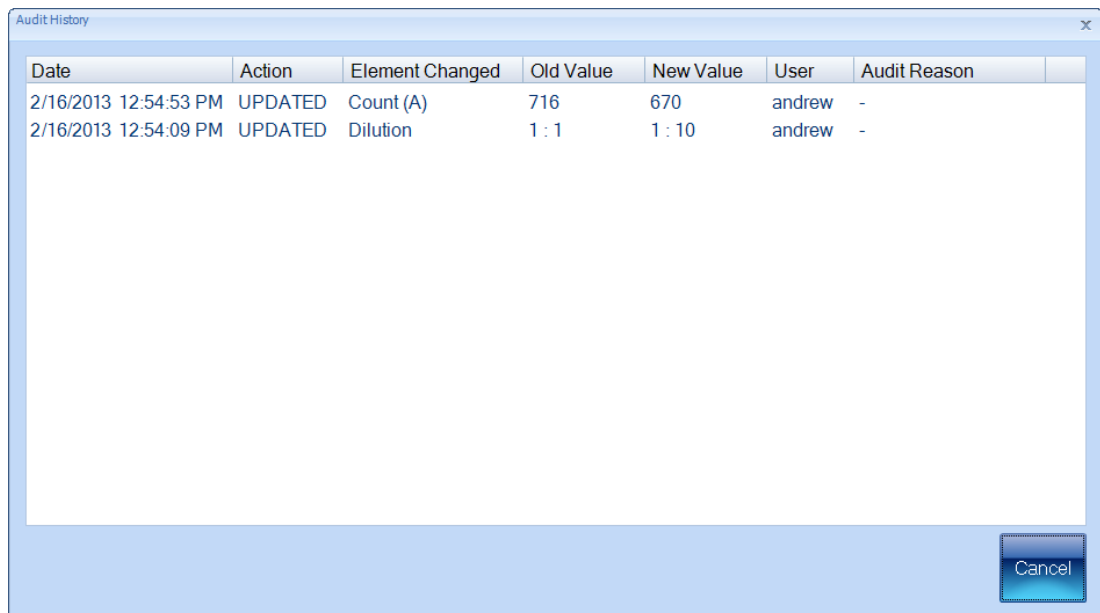
When you make any changes to a result, aCOLyte3 makes an entry in the audit history for that plate.

To view the audit history for a plate in the Results table:

1. Select the **Edit Result** tab for the plate result – see *Editing plate measurements*, page 78.
2. Press



to display the **Audit History** dialog box:



3. When you have finished viewing the audit history, press **Cancel** to close the dialog box.

### Manually adding and removing colonies

To add colonies to or remove colonies from the detected colonies for a result:

1. Select the **Edit Result** tab for the plate result – see *Editing plate measurements*, page 78.
2. If you want to remove all colony markers currently on the image so that you start the count from scratch, press



3. If you want to add colonies, press



If you want to remove colonies, press



4. If you have chosen to add colonies, click on the image at the point you want to add the colony – you cannot add colonies outside the frame.

If you have chosen to remove colonies, click on the colony markers you want to remove.

The count will be updated in the **Results** box as you click in the image.

5. Save the change to the result (see *Saving changes to results*, page 82).

The count(s) will be updated in the result together with any values depending on the count(s). An **M** flag will appear in the **Flags** column showing you have manually added or removed colonies.

## Adjusting the frame for a result

---

**Note** You cannot adjust the frame for a Spiral Plate batch.

---

To adjust the frame for a Pour Plate result:

1. Select the **Edit Result** tab for the plate result – see *Editing plate measurements*, page 78.
2. Press



3. Adjust the frame using the same techniques as when you create a batch or before taking a measurement – see *Adjusting the position and size of frames*, page 28.
4. Press



again.

The plate will be remeasured using the adjusted frame.

5. Save the change to the result (see *Saving changes to results*, page 82).

## Rejecting small particles in a result

When you create a batch, you can adjust the setting of the **Reject Small Particles** filter to remove the effect of small colored regions of the image that may be wrongly interpreted as colonies – see *Total Plate Count*, page 30. You can also change the **Reject Small Particles** filter in **Measurement Mode** before taking a measurement (see *Test measurement and adjusting settings*, page 72)

This section shows you how to change the **Reject Small Particles** setting for an individual plate *after* it has been measured.

To change the **Reject Small Particles** filter setting for an individual plate result:

1. Select the **Edit Result** tab for the plate result – see *Editing plate measurements*, page 78.
2. Drag the



slider; click the arrow buttons at the ends of the slider to adjust the slider by a single step; or press anywhere on the slider bar to set the slider to that position.

The plate will be remeasured with the new setting.

3. Check that the real colonies are still being counted while the small particles you want to reject are being excluded, and repeat Step 2 if the result is still not satisfactory.

---

**Note** You may find it helpful to zoom the image – see *Zooming the image*, page 10.

---

4. Save the change to the result (see *Saving changes to results*, page 82).

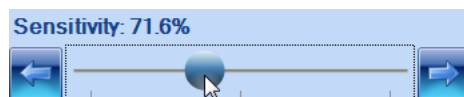
### Changing the Sensitivity setting for a result

When you create a batch you can adjust the detection **Sensitivity** – see *Total Plate Count*, page 30. After you have accepted the batch, you can also change the **Sensitivity** setting in Measurement mode *before* taking a measurement (see *Test measurement and adjusting settings*, page 72).

This section shows you how to change the **Sensitivity** setting for an individual plate *after* it has been measured.

To change the detection **Sensitivity** setting for an individual plate result:

1. Select the **Edit Result** tab for the plate result – see *Editing plate measurements*, page 78.
2. Drag the



slider; click the arrow buttons at the ends of the slider to adjust the slider by a single step; or press anywhere on the slider bar to set the slider to that position.

The plate will be remeasured with the new setting.

3. Check that the real colonies are being counted and the background and any debris are being excluded, and repeat Step 2 if the result is still not satisfactory.

---

**Note** You may find it helpful to zoom the image – see *Zooming the image*, page 10.

---

4. Save the change to the result (see *Saving changes to results*, page 82).

### Splitting colonies in a result

When you create a batch, you can choose whether to use the colony splitter to split overlapping colonies so that they are counted separately – see *Total Plate Count*, page 30. You can also change the colony splitter setting in **Measurement Mode** *before* taking a measurement (see *Test measurement and adjusting settings*, page 72).

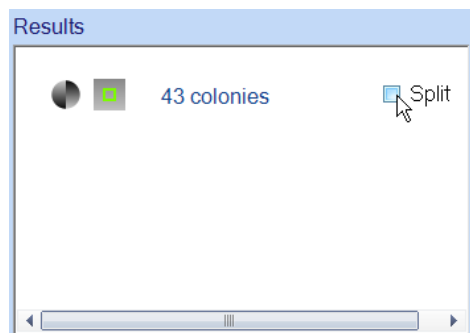
## Working with results

---

This section shows you how to change the splitter setting for an individual plate *after* it has been measured.

To change the colony splitter setting(s) for an individual plate result:

1. Select the **Edit Result** tab for the plate result – see *Editing plate measurements*, page 78.
2. Click on the **Split** check box in the **Results** box at the bottom of the **Edit Result** tab to change the setting:



The plate will be remeasured with the new split setting.

3. Check that overlapping colonies are being split satisfactorily and single colonies are not being split.

---

**Note** You may find it helpful to zoom the image when checking the effect of the colony splitter – see *Zooming the image*, page 10.

---

4. Save the change to the result (see *Saving changes to results*, page 82).

## Adding exclude regions to a result

---

**Note** You cannot place exclude regions on Spiral Plate batch results.

---

If required, you can add exclude regions to a plate image before you take a measurement from it – see *Exclude regions*, page 69. However, you can also add and remove exclude regions after you have performed the measurement – when you remove exclude regions after the measurement, you can clear exclude regions that were added before the measurement as well as those added to the result after the measurement.

To define exclude regions for a plate result:

1. Select the **Edit Result** tab for the plate result – see *Editing plate measurements*, page 78.
2. Press



to select it:



---

**Note** You may find it helpful to zoom the image before placing the exclude region on the image – see *Zooming the image*, page 10.

---

- You define the exclude region in the same way as when you add exclude regions before making a measurement – see *Exclude regions*, page 69.

The plate will be remeasured with the new exclude region in place.

- To remove the last exclude region you added to the image (you can repeat the procedure to remove each of the regions in turn, including any added before the measurement was made):

Press



The plate will be remeasured with the last exclude region removed.

- Save the change to the result (see *Saving changes to results*, page 82).

### Zeroing the count

If the result for a plate is faulty in some way and you wish to exclude it, you can zero the count. If you zero a result, it will still be shown in the Results table (see *The Results table*, page 77) with count(s) equal to 0.

**Notes** This section shows you how to set the count(s) for a result to zero to record a zero result.

If you just want to remove all the colony markers from the image and set the counts to zero so that you can count the colonies manually, press **Clear Markers** – see *Manually adding and removing colonies*, page 83.

If you want to delete a faulty result, press **Delete Plate** – see *Deleting results*, page 82. However, if you delete a result, it will be removed from the results table completely, and you will not be able to reset the change later if you change your mind about excluding the result (see *Undoing and resetting changes*, page 82).

To set the count(s) for a plate to zero:

- Select the **Edit Result** tab for the plate result – see *Editing plate measurements*, page 78.
- Press



All other result editing controls will be disabled apart from **Save Changes**, **Undo All**, **Reset** (if there are previously saved changes) and the controls in the **Editing Plate** panel at the bottom of the aCOLyte3 window.

- Save the change to the result (see *Saving changes to results*, page 82).

The result will be zeroed in the Results table.

**Note** When you zero a result that contributes to a mean, the result will still contribute to the mean (with count 0). If you want to remove the result's contribution to the mean, you can change its plate identifier (see *Editing the plate identifier, comments and dilution*, page 81) or delete the result (see *Deleting results*, page 82).

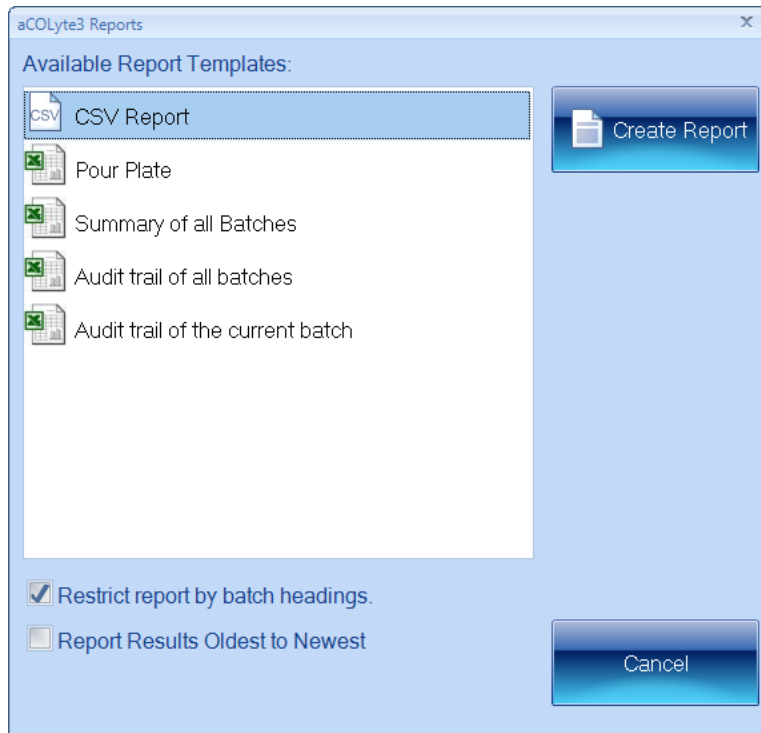
### Compiling reports

To compile a results report:

1. Press



in the **Results** tab to display the **aCOLyte3 Reports** dialog box:



2. Click on the template for:
  - **CSV Report** to save a report on the current batch including system settings to a CSV file
  - the current batch (labeled by the batch type) to create a detailed report for the current batch
  - **Summary of all Batches** to create a summary report covering all the batches in the aCOLyte3 database
  - **Audit trail of all batches** to create an audit report covering all the batches in the aCOLyte3 database
  - **Audit trail of the current batch** to create an audit report for the current batch.
3. To limit the information given for each result in the current batch report to the columns currently shown in the Results table (the default):
  - Leave **Restrict report by batch headings** checked.

To include all the data for each result, including columns not selected for the Results table (see *The Results tab*, page 54):

Click **Restrict report by batch headings** so that it is not checked.

---

**Note** The **Restrict report by batch headings** setting has no effect on summary or audit trail reports.

---

4. Press



5. If you chose to create a CSV report, a dialog box will be displayed showing you the name of the report – press **OK** to close the dialog box.

Otherwise, Microsoft Excel will open with a worksheet containing the selected report.





# Configuring aCOLyte3

---

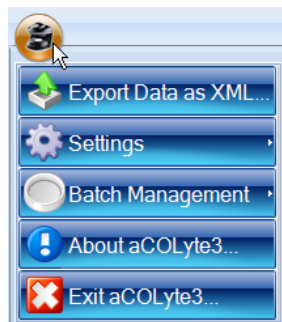
This chapter shows aCOLyte3 administrators how to configure aCOLyte3 in a variety of ways. You will find out about:

- Displaying information about the program version you are using – see *About aCOLyte3*, below
- Using the **Settings** dialog box to configure the program – see *System settings*, on the next page
- Changing the color of the frame – see *Managing aCOLyte3 users*, page 95
- Setting up user accounts – see *Managing aCOLyte3 users*, page 95
- Specifying the location of the aCOLyte3 database on the PC or a network– see *Database location*, page 100
- Calibrating the instrument – see *Calibration*, page 101.

## About aCOLyte3

To display information about the version of aCOLyte3 you are running:

1. Press the aCOLyte3 button at the top left-hand corner of the window to display the aCOLyte3 menu:



2. Press



## Configuring aCOLyte3

---

to display the **About aCOLyte3** dialog box:

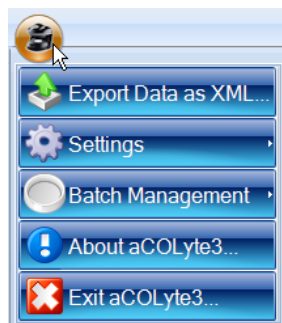


3. Press **Save Details** to display a standard Windows **Save As** dialog box if you want to save the program version details in a text file.
4. Press **Close** to close the dialog box.

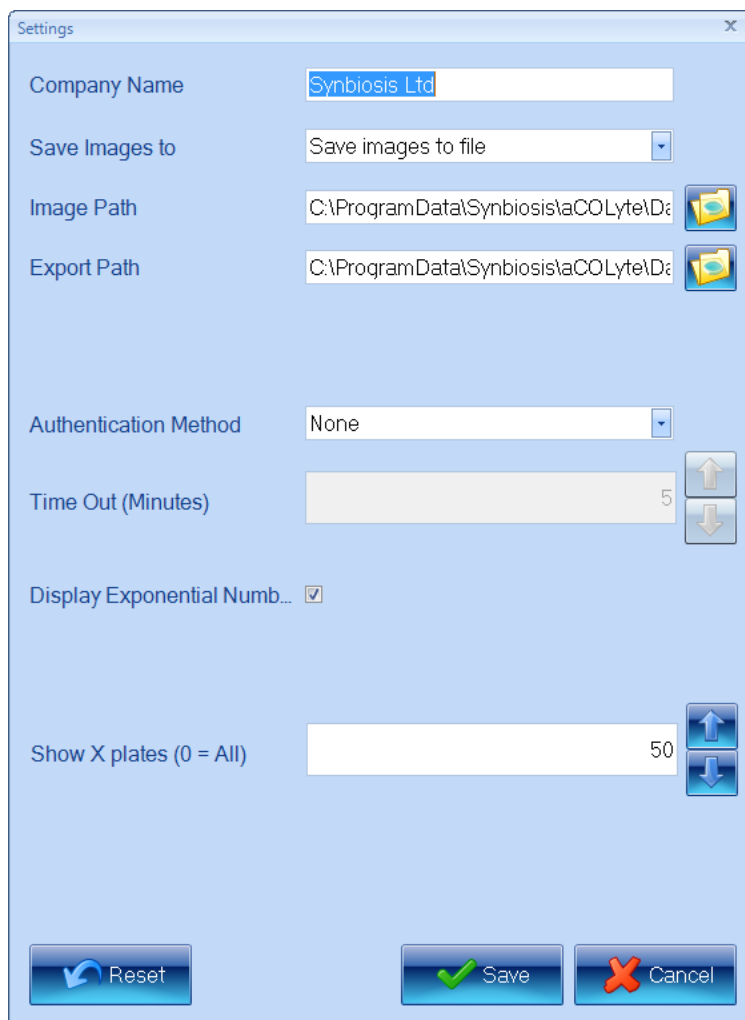
## System settings

To view or change the system settings:

1. Press the aCOLyte3 button at the top left-hand corner of the window to display the aCOLyte3 menu:



- Choose **System Settings** from the **Settings** submenu to display the **Settings** dialog box:



- Enter the name of your organization under **Company Name**.  
The **Company Name** is included in reports – see *Compiling reports*, page 88.
- In the **Save Images to** box, choose whether to **Save Images to file** when you carry out a measurement; choose **Do not save images** if you do not want the images saved.

---

**Note** You will only be able to edit results after you have performed a measurement if the image is saved with the results – see *Editing plate measurements*, page 78, for details.

---

- Set the **Image Path** for the folder used to save images by typing the path directly into the box, or press



to display a standard Windows **Browse For Folder** dialog box so that you can select the required folder.

## Configuring aCOLyte3

---

6. Set the **Export Path** for the folder used to save exported batch details (see *Exporting batch details*, page 57) by typing the path directly into the box, or press



to display a standard Windows **Browse For Folder** dialog box so that you can select the required folder.

7. Select the **Authentication Method** you want to use – you can choose:

- **None**
- **Windows**
- **Local User Groups**
- **Network User Groups.**

See *Managing aCOLyte3 users*, on the facing page, for full details of what these mean and for how to set up Local and Network User Groups.

8. Press



to reset all settings to the values they had when you opened the dialog box.

9. Press

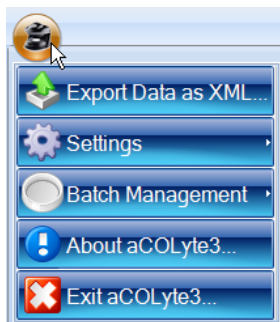


to confirm the new settings and close the dialog box.

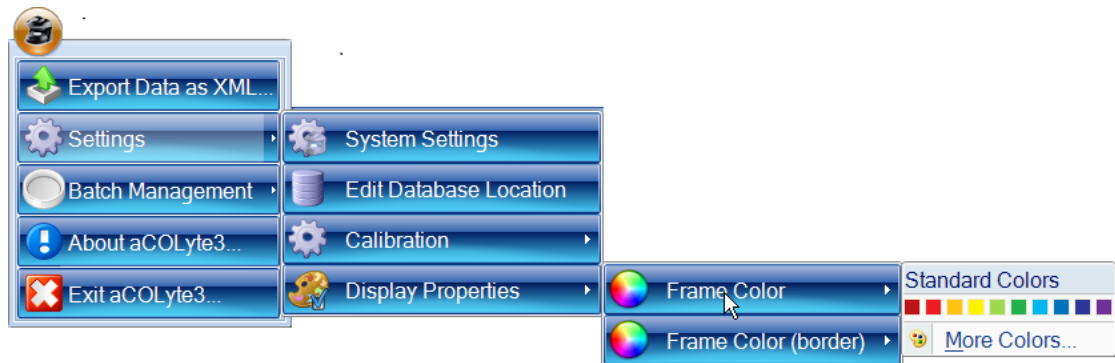
## Display properties

To set the color of the frame or frame border:

1. Press the aCOLyte3 button at the top left-hand corner of the window to display the aCOLyte3 menu:



- Choose **Frame Color** or **Frame Color (border)**, from the **Settings→Display Properties** submenu to display the Color palette, for example:



- Either click on one of the **Standard Colors** in the palette to select it or press **More Colors** to display a color picker allowing you to choose from a wider range of **Standard** colors or to create your own **Custom** color.

**Note** The border color only applies to spiral frames. In particular, it is the color of the parts of the spiral that are not included in the two sectors of a two-sector frame, and it is the color of the inner boundary of an annular frame.

## Managing aCOLyte3 users

You use the **Settings** dialog box (see *System settings*, page 92) to specify the **Authentication Method** used by aCOLyte3 to control access to the program. The options are:

- **None** – in this case there is no access control and:
  - anybody will be able to use aCOLyte3 without having to log on to the program.
  - audit trails will record that changes were made by the user who was logged in to Windows at the time the change was made.

If you choose **None** as the **Authentication Method**, there is nothing more to do after you have made the selection.

- **Windows** – in this case:
  - users will have to log on to aCOLyte3 using their Windows user name and password (it does not have to be the user currently logged on to the PC, provided they have a user name and password to log on to that PC)
  - audit trails will record that changes were made by the user who was logged in to aCOLyte3 at the time the changes were made.

If you choose **Windows** as the **Authentication Method**, you will need to add user names and passwords to Windows for all the users who are going to use the aCOLyte3 installation – see *Windows password security*, on the next page.

- **Local User Groups** – In this case:
  - users must be members of the **Acolyte Users** local user group on the PC
  - users will have to log on to aCOLyte3 using their Windows user name and password (it does not have to be the user currently logged on to the PC, provided they are able

## Configuring aCOLyte3

---

to log on to that PC)

- audit trails will record which aCOLyte3 user made any changes.

If you choose **Local User Groups**, you will need to:

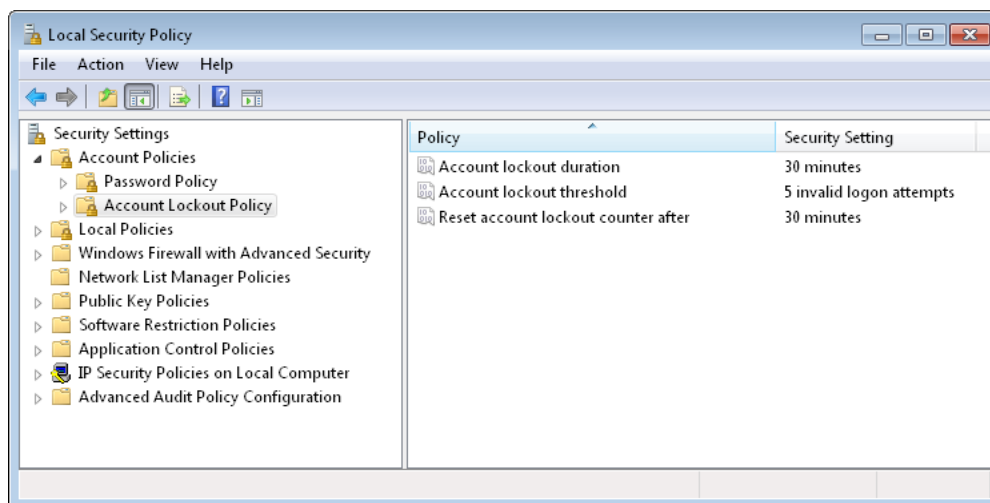
- add Windows user names and passwords for all the users who are going to use the aCOLyte3 installation – see *Windows password security*, below
- define the **Acolyte Users** local user group on the PC – see *aCOLyte3 user groups*, on the facing page
- **Network User Groups** – this is the same as the previous option, except the **Acolyte Users** user group is defined at the Network level – see the same set up references as for **Local User Groups**.

For details of how to view audit trails, see *Viewing the audit history of changes*, page 83.

## Windows password security

Unless you choose to allow completely free access to the program (**Authentication Method** set to **None** in the **Settings** dialog box – see *System settings*, page 92), aCOLyte3 uses Windows password authentication to control access. This means that in order to log on to aCOLyte3, the user must enter a valid Windows user name and password for the PC – note that it does not have to be the user name and password of the currently logged on Windows user.

The fact that aCOLyte3 uses Windows password authentication also means that the password security policy is determined by Windows, and can be set by the Windows administrator. For example, for a stand-alone aCOLyte3 system, running secpol.msc displays the **Local Security Settings** window:



These settings determine the password policy for aCOLyte3 on the PC, so, for example, setting the **Account lockout threshold** to **5 invalid logon attempts** would mean that if the user made five invalid log on attempts, nobody will be able to log on to aCOLyte3 on the PC during the **Account lockout duration**.

### aCOLyte3 user groups

If the **Authentication Method** is set to **Local User Groups** or **Network User Groups** in the **Settings** dialog box (see *System settings*, page 92), in order to gain access to aCOLyte3, a user must:

- a. have a valid user name and password to log on to Windows on the aCOLyte3 PC – see *Windows password security*, on the previous page, for further details
- b. be a member of the **Acolyte Users** local or network user group – see the next section, *Local and Domain/Security groups* for details.

### **Local and Domain/Security groups**

There is a single aCOLyte3 user group: **Acolyte Users**.

You can set up the **Acolyte Users** group using:

- **Local groups** on each aCOLyte3 PC.

Generally speaking, you would choose to use Local groups if you have one or more independent aCOLyte3 installations. You will need to set up the **Acolyte Users** group separately on each of the aCOLyte3 PCs – see the next section, *Local groups*.

Or:

- **Domain/Security groups** defined across a Windows Server domain.

If the aCOLyte3 PCs are connected to a network arranged in a Windows Server domain, you can use Windows Domain/Security groups to define the **Acolyte Users** group. In this case the user group is set up centrally on a domain controller rather than on the individual aCOLyte3 PCs – see *Domain/Security groups*, page 100.

When the aCOLyte3 PCs are arranged in a Windows Server domain network, it is also usually convenient for them all to be connected to a central aCOLyte3 database server rather than having separate independent databases on each PC – see *Database location*, page 100, for how to link a aCOLyte3 installation to a central database.

The following sections show you how to set up and use *Local groups* and *Domain/Security groups*, page 100.

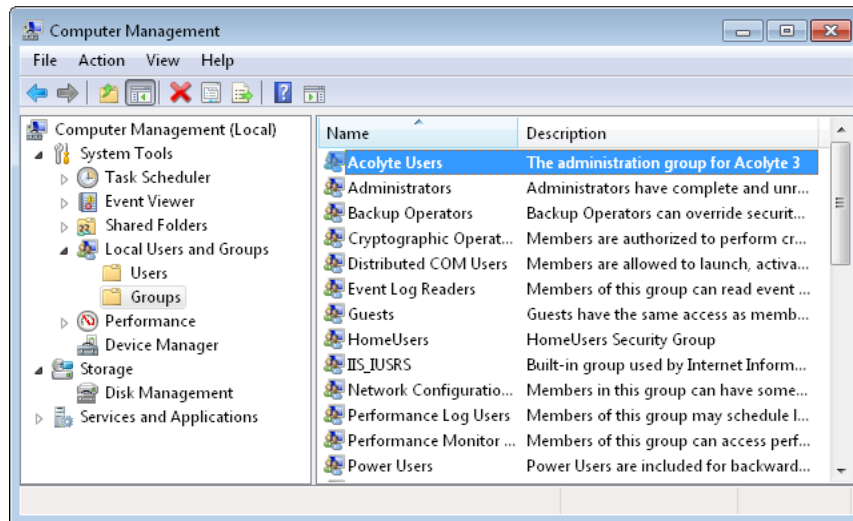
### **Local groups**

You need to set up Local groups separately on each aCOLyte3 PC – see *Domain/Security groups*, page 100, for how to set up Domain/Security groups for use across a network.

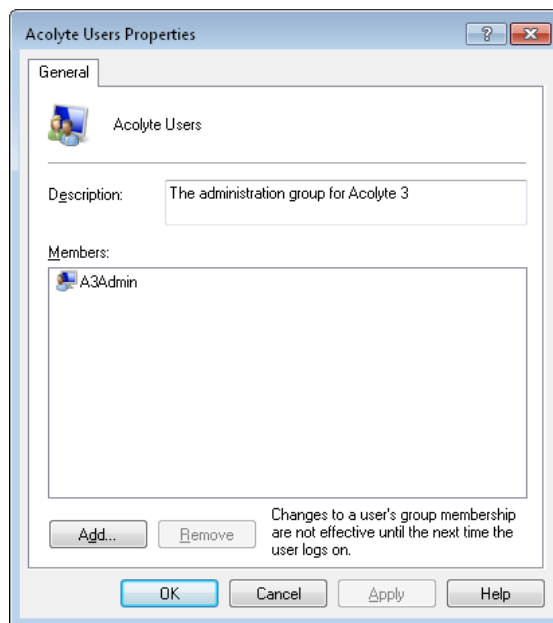
To set up Local user groups:

1. Open the Windows Control Panel.
2. Open the **Administrative Tools**.

3. Open **Computer Management**, expand **Local Users and Groups** and select **Groups**:



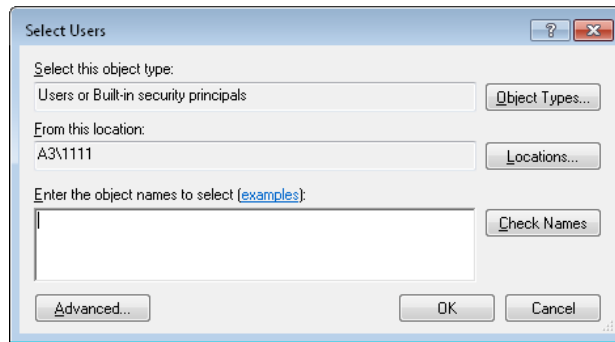
4. To add users to the **Acolyte Users** group:
  - a. Right-click on the required group in the right-hand pane to display a context menu.
  - b. Choose **Add to Group** to display the **Group Properties** dialog box:



The example shows the default **aCOLyte Users** group, which contains the **A3Admin** user by default.



- c. Press **Add** to display the **Select Users** dialog box:

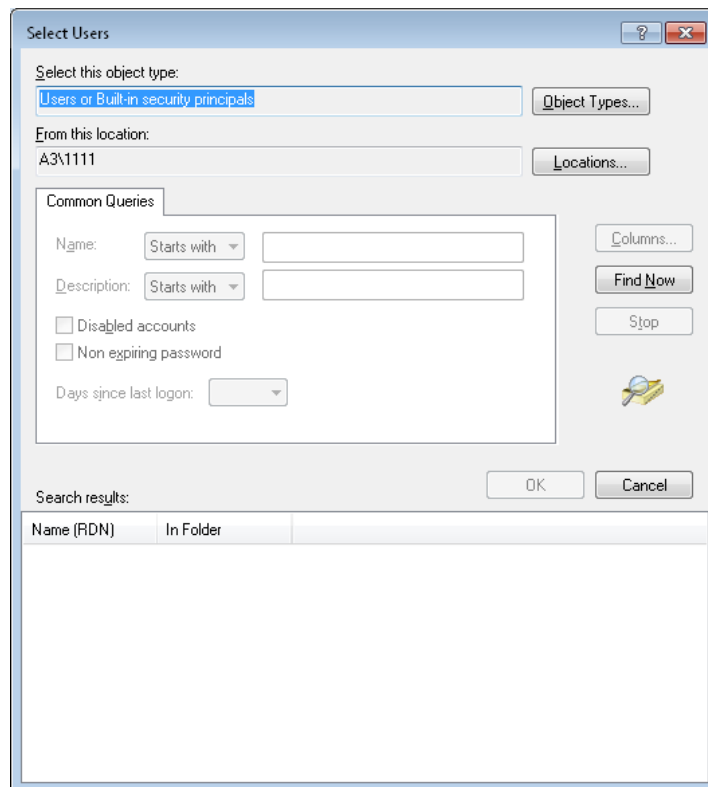


Either:

- i. Type the user name into the box; if required, you can enter multiple user names by typing them on the same line but separated by semi-colons.
- ii. Press **Check Names** to check that the user names are valid Windows log on user names – if a user name is not found, and you entered it correctly, you will need to create a new user account with this user name.

Or:

- i. Press **Advanced** to expand the **Select Users** dialog box:



- ii. Press **Find Now** to list the Windows users for this PC.
- iii. Select the required users to add to the group.
- iv. Press **OK** to close the dialog box and add the users to the box in the simple **Select Users** dialog box.

## Configuring aCOLyte3

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- d. Press **OK** to close the **Select Users** dialog box and add the users to the group in the **Properties** dialog box.
- 

**Note** If required, you can also use the **Properties** dialog box to remove users from the group by selecting them and pressing **Remove**.

---

- e. Press **OK** to close the **Properties** dialog box.

### Domain/Security groups

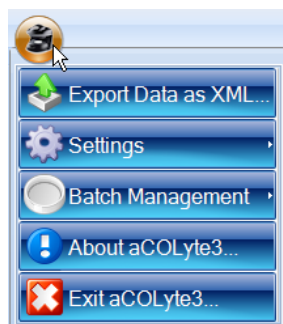
The principles and procedures for creating and populating an **Acolyte Users** Domain/Security group are similar to those described for *Local groups*, page 97, but using the Active Directory Users and Computers console instead of the Computer Management application.

## Database location

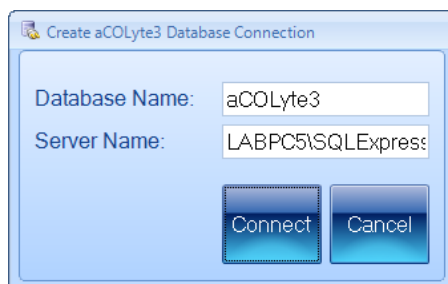
If you are using several aCOLyte3 instruments connected across a network, they can share a single aCOLyte3 database. In order to do this, you will need to specify the location of the shared database in each of the aCOLyte3 installations. Alternatively, you may have multiple databases on the local database server on your PC, in which case, you can use the following procedure to specify which one to use.

To specify the location of the aCOLyte3 database:

1. Press the aCOLyte3 button at the top left-hand corner of the window to display the aCOLyte3 menu:

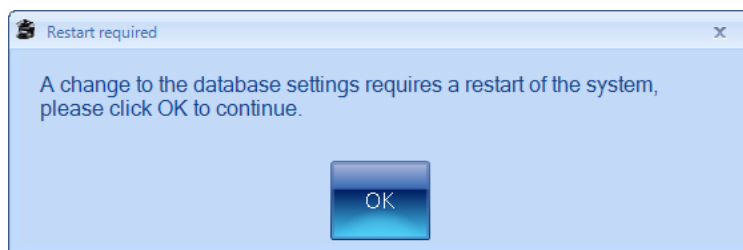


2. Choose **Edit Database location** from the **Settings** submenu to display the **Create aCOLyte3 Database Connection** dialog box:



3. Enter the names of the aCOLyte3 **Database** and **Server**.
4. Press **Connect**.

A dialog box will be displayed warning you that aCOLyte3 will restart:



5. Press **OK**.

aCOLyte3 will close down and restart automatically connected to the new database location.

## Calibration

aCOLyte3 is fully calibrated in the factory and in normal circumstances will require no further calibration after delivery. However, in exceptional circumstances you may need to carry out some recalibration. For details, see:

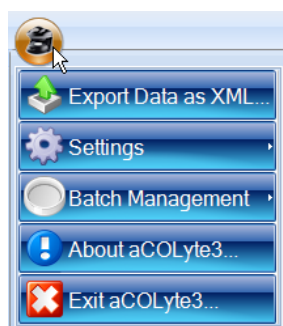
- *Camera measurements*, below.

### Camera measurements

aCOLyte3 is fully calibrated in the factory and in normal circumstances will require no further calibration after delivery. However, in exceptional circumstances you may need to recalibrate the camera measurements, which enable the instrument to measure distances on the image accurately.

To recalibrate the camera measurements:

1. Insert an object of known size into aCOLyte3 – see *Loading plates into the instrument*, page 8.
2. Press the aCOLyte3 button at the top left-hand corner of the window to display the aCOLyte3 menu:



3. Choose **Calibrate Camera Measurements** from the **Settings**→**Calibration** submenu to display the **Calibrate Camera for Measurements** dialog box.

This dialog box is also displayed when you calibrate an imported image, and is used in exactly the same way – see *Calibrating the image*, page 12, for detailed instructions.



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