# **Gallios** <sup>TM</sup> **Quick Reference**

## **Purpose:**

The purpose of this Quick Reference is to provide a simple step by step outline of the information needed to perform various tasks on the system. We begin with basic tasks like starting up or shutting down the system and then proceed to more involved software tasks. It is organized according to the modules in the Gallios Flow Cytometer Training Modules. Major Headings (in blue) are the major groups of tasks. These headings may be further subdivided into subsections of the major task and appear in a logical sequence. For example, the Major task of creating a protocol is subdivided into the subsections: Select Parameters, Create Plots, Instrument Settings, Create Regions, Gate, Analyze, and Save Protocol. A list of the major tasks appears below in the Table of Contents.

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#### **Startups**

- Check external Sheath tank, fill the Cleaning agent tank and Empty the Waste tank
- Power the Computer and Monitor On
- Enter a Windows password if any and select **OK**



- Double click Software icon.
- Select User name
- Enter User password and select Next
- Select **Connect** (to run samples) or **Work Offline**.
- Check gages System Pressure 30 Psi, Vac 17 in (Cytometer only)



#### **Commonly Used Toolbars:**





- Place 2 ml of 10 % bleach solution in one tube
- Place 2 ml of de-ionized water in each of 3 tubes .
- on the toolbar to clear the Acq. Mgr. Select Select the **tab** on the Resource Explorer
- Select the Common Folder
- Select the **cleanse.pnl** and drag and drop it to the Acq. Mgr.
- Enter a carousel number
- button Select
- Select Yes if you wish to save the previous protocol or No if not
- When the panel is finished, remove all tubes .
- Select <sup>ZZZ</sup> Standby mode
- When finished select  $\times$  in the upper right to exit the . software
- Select No



- Select Cytometer Off to turn off the cytometer
- Select **Start** >> **Shutdown** >> **OK** to shutdown the computer

# **Creating a Protocol**

Select Parameters:	<ul> <li>Select File &gt;&gt; New &gt;&gt; New Protocol to clear the workspace of all but one dotplot and single elliptical gate region.</li> <li>Select Yes to save old protocol or No if you do not wish to save it</li> <li>Select I Reference Control on the toolbars</li> <li>Select Acquisition Setup tab</li> <li>Set Limits and Max events to stop accumulation later</li> <li>Select I Parameters.</li> <li>Select unwanted parameters</li> <li>Select any desired parameters</li> <li>Optional: Set Ratio, Time and Derived Parameters</li> <li>Close shutters of unused lasers</li> <li>Select FS Collection angle and SS desired ND filter</li> <li>Select OK</li> <li>Select Yes, enter a name and select Save</li> </ul>
Create Plots:	<ul> <li>Dot Plots (2 parameter) – Select , Histogram Plot (Single parameter)– Select , Density plot – Select </li> <li>Select Parameter(s) and then OK</li> <li>Repeat for each plot</li> <li>Select each plot Focus (top of the plot) in turn and drag the plot to the desired location on the Workspace</li> <li>Press CTRL + T, select plot size, and then OK to arrange plots</li> </ul>
Instrument Settings:	<ul> <li>Select Select Run</li> <li>Select Cytometer Control and then the Acq. Setup tab</li> <li>Select Setup Mode and QuickSET</li> <li>Choose discriminator parameter and adjust Discriminator to exclude debris and noise but pass cells</li> <li>Adjust the slider bars (top and side of each plot) beginning with those to be used as gates for a correct pattern</li> <li>Select Acq. Setup tab</li> <li>If needed, select QuickCOMP and adjust the compensation using a proper sample until correct patterns are seen</li> </ul>

	<ul> <li>Deselect Setup Mode, QuickSET, and QuickCOMP</li> <li>Allow the system to collect the desired amount of data</li> <li>Select to stop or allow the system to reach a stop value</li> </ul>
Create Regions	<ul> <li>Select each plot in turn and add regions:</li> <li>Dot Plots (2 Parameter Plots only): <i>Polygon</i> - select  <ul> <li>Move the cursor to starting point on plot and left click Use a click move technique to create all the sides back to the starting point</li> <li><i>Rectangle</i> - select  </li> <li>Move the cursor to the starting corner and left click Move the cursor to the opposite corner and left click Move the cursor to the opposite corner and left click Move the cursor to plot and left click Move the cursor to plot and left click Move the cursor to plot and left click Move the region to anchor position and left click Move the region to anchor position and left click Move the cursor to desired population and left click</li> </ul> </li> <li>Histogram Plots:</li> </ul>
	Linear – Select Move the cursor to starting position and left click Move the cursor to ending position and left click Multilinear – Select Move the cursor to first position and left click Move the cursor to end of first region and left click Move the cursor to end of second region and left click Move the cursor to end of second region and left click Continue until all regions have been created Move cursor to Move cursor to Move the click to finish
Gate	<ul> <li>Move the cursor to lower right corner of plot to be gated</li> <li>When appears, left click and select Gate</li> <li>If you wish to apply the gate to all plots select that option</li> </ul>
Optional: Stop Count	<ul> <li>Select Stop and Save tab and enter count</li> <li>Select OK</li> </ul>
Analyze	<ul> <li>Select Analysis &gt;&gt; Select Results</li> <li>Deselect all unwanted statistics and select all desired statistics and Select OK</li> </ul>
Save Protocol	<ul> <li>Select File &gt;&gt; Save Protocol or Select II</li> </ul>

### **Create a FlowPAGE**

	<ul> <li>Select Insert &gt;&gt; Blank Flow Page</li> <li>Move the new page to a convenient area of the screen</li> </ul>
Add Plots	<ul> <li>Move the cursor to the center of a plot</li> <li>Left click and drag and drop it onto the FlowPAGE</li> <li>Repeat for each plot</li> </ul>
Arrange Plots	<ul> <li>Select the Maximize Window icon in the upper right corner of the FlowPAGE to expand the page to full screen Move each plot and move into position OR</li> <li>Create a large rectangle completely encircling two plots and stats</li> <li>Select the appropriate alignment tool (<sup>PP</sup> top, <sup>bb</sup> bottom, <sup>bb</sup> left or <sup>*D</sup> right)</li> </ul>
Add Text	<ul> <li>Select A on the FlowPAGE toolbar</li> <li>Move the cursor to the location on the page and left click</li> <li>Type in desired text and select OK</li> <li>Reshape the text box by grabbing a handle and pulling outward</li> <li>Right click in the box and select Properties to change font, fill, etc.</li> <li>Left click on another part of the page to finish</li> </ul>
Add Images	<ul> <li>Select an on the FlowPAGE toolbar</li> <li>Locate the directory where the image is stored</li> <li>Select the desired image and then <b>Open</b></li> <li>Move the cursor to the desired location on the page and left click</li> <li>Use the handles to resize the image</li> <li>Left click on another part of the page to finish</li> </ul>
Add Lines or Boxes	<ul> <li>Select or </li> <li>Move to the starting point</li> <li>Hold the left button and drag to the ending point</li> </ul>
Save FlowPAGE	Select FlowPAGE >> Save FlowPAGE As Enter a name and select Save

# AutoSetup

Note: To enter a new application, you will need a base protocol and target channels for Flow-Set Pro

New Application	Select Tools >> AutoSetup Application Definition Select Create a new application definition Select Current user only or All users as desired Select Next Browse to find the base protocol that represents the application Select the base protocol and then <b>Open</b> Select Next Select Next Select Filter Block and then Next Optional: select dyes Select Next Enter the correct Flow-Set Pro target channels Select Next Enter an application name and select Next Select Create a verification protocol if you wish the system to generate a verification protocol if you wish the system to generate a verification protocol Or Select Next The system summarizes your entries Select Print if you wish and then Finish The system now creates all protocols and FlowPAGES needed for Autostandardization
Schedule an Application	Select Tools >> AutoSetup Scheduler Select desired application Or Hold the CTRL key down and select multiple applications Enter a carousel number and select Schedule Select Run if you wish to run immediately Or Select Close to prep tubes, place them on the system, or enter a calibration factor in the Acquisition Manager Select I in the toolbar to run the samples

# **Acquisition Manager (Panels)**

	<ul> <li>If it is not visible, select View (or right click)</li> <li>Select Acquisition Manager</li> </ul>
Select Columns	<ul> <li>Select View &gt;&gt; Customize Worklist Columns</li> <li>Select the desired columns to appear and then OK</li> </ul>
Edit Column Names	<ul> <li>Select View &gt;&gt; Customize Worklist Columns</li> <li>Left click on the column name until the name can be changed</li> <li>Type in the new name and press Enter</li> <li>When finished, select OK</li> </ul>
Create a Panel	<ul> <li>Select is to clear the Acquisition Manager</li> <li>Select is protocol tab on the Resource Explorer</li> <li>Drag and drop the desired first protocol to the Acquisition Manager</li> <li>Select the is on the toolbar to add the desired number of tubes to the panel</li> <li>If you wish to change the protocol in any of the tubes, select the is Open Protocol in the Protocol column for that tube</li> <li>Select the desired protocol and then <b>Open</b></li> <li>Repeat as necessary for each tube</li> <li>Under Regions column, toggle the appropriate icon: to use the regions in that protocol, to carry the region to the other tubes, or to import regions from another protocol</li> <li>Under the Cytosettings column, toggle the appropriate icon to: to use the cytosettings in the protocol, to pass the cytosettings to the next protocol, or to pass the cytosettings to the next protocol, or Select the actual measurements (example: CD3, CD4, etc.)</li> </ul>
To Delete a Protocol	• Select the protocol line by its number and press <b>Delete</b>
To Move a Protocol	<ul> <li>Select the protocol and drag and drop to the new position</li> </ul>

Save Panel	•	Right click on Panel folder or panel area in Acq. Mgr. Enter a panel name If you desire to create a template in the database for Reporting results, select Export results of panel to Report Generator on the Panel Save Dialog Select <b>Save</b>
Create a Worklist	-	Select Clear Worklist to clear the Acq. Mgr. then:
To Add Panels		Select Panels tab on the Resource Explorer Locate the desired panel and drag and drop it to the Acquisition Manager Repeat as desired
To Add Protocols	•	Select Protocols tab on the Resource Explorer Locate the desired protocol and drag and drop it to the Acquisition Manager Repeat as desired
Editing Parameter Names	:	Select the desired field (e.g. Under P1, P2, etc) Type in the desired information (e.g. Sample ID, Parameter name, etc.)
Save Worklist	:	Select Save Worklist on the toolbars Enter a worklist name and select <b>Save</b>
To Run		Select Select Worklist tab on Resource Explorer Locate the desired worklist and drag and drop it to the Acquisition Manager Edit the boxes as desired To add Cal Factor, click on I in the Cal Factor column of the Acquisition Manager Enter absolute count calibration value, verify Enabled has been selected, and select <b>OK</b> Enter a carousel number in tube one position Select Run

## DataBase

QC Product Editor	
Editing a Product	<ul> <li>From the Report Generator toolbar select Product Editor</li> <li>Select the desired product you wish to edit</li> <li>Select the desired information field to be edited</li> <li>Make the change</li> <li>Repeat the process for each field</li> <li>When finished, select Save Prod then select Yes to confirm update to the lot number</li> <li>Select OK</li> </ul>
Entering a New Product	<ul> <li>From the Report Generator toolbar select</li> <li>Product Editor</li> <li>Select the New Product button</li> <li>Select the first information field</li> <li>Enter the information</li> <li>Repeat the process for each field</li> <li>When finished, select Save Prod then select Yes to confirm update to the lot number</li> <li>Select OK</li> </ul>
Quality Control Report	Sample data is sent to the database when samples are run
Setting Region Properties	<ul> <li>and the protocol region properties are set correctly</li> <li>Drag and drop a protocol to the workspace</li> <li>Save the previous protocol or not as desired</li> <li>Select a region for statistics to be sent to the database</li> <li>Right click the mouse and select <b>Region Properties</b></li> <li>Select Region statistics to be exported for Quality Control</li> <li>Select a QC Product and select <b>OK</b></li> <li>Repeat for each region to be used</li> <li>Save the protocol</li> </ul>
Activating QC Template (one time procedure)	<ul> <li>Select on the Report Generator toolbar</li> <li>Change instrument to Default</li> <li>Select Template &gt;&gt; Select Template</li> <li>Select desired template and then Open</li> <li>Select Template &gt;&gt; Save Template to save the template with the system serial number</li> <li>Repeat process for each template</li> </ul>

Select a Template	<ul> <li>Select on the Report Generator toolbar</li> <li>Select Template &gt;&gt; Select Template</li> <li>Select desired template and then Open</li> </ul>
Select a point	• Left click on the point (a red line appears)
Creating a New QC Template	<ul> <li>First, enter the new QC product (earlier in database) and ensure the protocol has regions selected for QC export with a QC product</li> <li>Run the protocol at least once</li> <li>Select I I I I I I I I I I I I I I I I I I I</li></ul>
Panel Template	
Accessing a template	<ul> <li>Select in the Report Generator toolbar</li> <li>Select File &gt;&gt; Open and locate the desired template</li> <li>Select the template and then Open</li> </ul>
Creating a Template	<ul> <li>First create a panel and then save the panel with the</li> <li>Export results of panel to report Generator selected</li> <li>Select Panel Template on the Report Generator toolbar</li> <li>Select File &gt;&gt; Open and locate the new blank</li> </ul>

- template
  Select the template and then **Open**Select the desired global options on the Local tab

	<ul> <li>Select each field in turn and enter the desired information</li> </ul>
	<ul> <li>Select Update to enter the information and clear the fields</li> </ul>
	Or
	<ul> <li>Select Update &amp; Retain to enter the information only</li> </ul>
	<ul> <li>Repeat for each line</li> </ul>
	<ul> <li>Select Data Plots tab to add plots</li> </ul>
	When finished, select File >> Save
Panel Report toolbar	<ul> <li>Select Search button to list reports</li> <li>Select Search button to list reports</li> <li>Highlight desired report and then click on Select</li> </ul>
Database Entry	• Select on the Report Generator toolbar
	<ul> <li>Select New</li> <li>Enter information in each desired field and select</li> </ul>
	Save
Maintenance Log	<ul> <li>Select , double click on item to log</li> </ul>
Service Log	<ul> <li>Select 5<sup>(N)</sup>, select field and enter information</li> </ul>

#### **Offline Analysis**

Retrieve with Runtime Protocol (protocol used To collect the data)

Retrieve with a Different Protocol

Retrieve Multiple Listmode Files with Runtime Protocols

- Select Listmode tab on the Resource Explorer
- Locate the desired listmode file and drag and drop the file to the Workspace
- If asked do you wish to clear acquisition data, answer **Yes**
- Save previous protocol as desired
- The protocol appears and the data shortly after
- Select **B** Protocols tab on the Resource Explorer
- Locate the desired protocol drag and drop it the Workspace
- If asked do you wish to clear acquisition data, answer Yes
- Save previous protocol as desired
- Select Listmode tab on the Resource Explorer
- Locate the desired listmode
- Drag and drop it to any one of the plots on the Workspace
- The protocol will populate with the data
- Select Tools >> Listmode Playback
- Right click on Add Protocol tool in the window
- Select New Worklist
- Select Add Protocol tool once for each addition protocol
- Under the LMD File column, select Open File icon
- Select the desired listmode file and then **Open**
- Repeat for all listmode files
- Select the Options tab and other desired options
- Select the Single File Mode tab
- Select the Run tool in the window
- The files are displayed

Same as previous action but with one addition:

Files with Different Protocols

	<ul> <li>Select the Open Protocol icon under the Protocol column</li> <li>Select the desired protocol and then Open</li> <li>Repeat for each protocol</li> </ul>
Batch Automator	<ul> <li>Select Tools &gt;&gt; AutoMATOR Setup</li> <li>To remove old selected files from the list: select the files and then the Remove Files button</li> <li>Select Add Files</li> <li>Select the desired files and then Open</li> <li>To change the order: Select a file and drag and drop it to a new position</li> <li>Select the desired process option next to the files (↓ pass, A pause 12 seconds, ⊕ stop and wait to restart)</li> <li>Select the desired output option</li> <li>To process: Select</li> <li>Run on the Automator toolbar</li> </ul>
Listmode Playback Tool	<ul> <li>Select Tools &gt;&gt; Listmode Playback</li> <li>Select either the Add Protocol or Add Panel</li> <li>Select in Open for the first listmode file</li> <li>Select the listmode file and then Open</li> <li>Repeat for each listmode file</li> <li>If desired, select the Open for each protocol in turn</li> <li>Select the desired protocol and then Open</li> <li>Select Single File Mode tab</li> <li>Select in the window</li> </ul>
Publishing Set Preferences	<ul> <li>The files are displayed</li> <li>Press CTRL + W and select Publish tab</li> <li>Select the desired publishing option Publish to Text File or Publish Data to MS Excel or both</li> <li>Select desired data format</li> </ul>

Select desired bitmap size

	<ul> <li>Select desired File References option</li> <li>Select whether or not to switch to Excel on publish</li> <li>Select OK</li> </ul>
Output	<ul> <li>Select Tools &gt;&gt; Publish Results Now</li> <li>Enter name and select Save</li> <li>System opens MS Excel and displays data</li> </ul>
Assigning Color to Dot Plots	<ul> <li>Press CTRL + W and select Gating tab</li> </ul>
Color Precedence	<ul> <li>Select Automatic Color Precedence</li> <li>Select OK</li> <li>Select Color Precedence Setup on the toolbars</li> <li>Edit colors as desired</li> <li>Move regions using a drag drop technique</li> <li>Select OK</li> <li>Color will be assigned to dots based on the color located highest on Precedence list (top position)</li> </ul>
Color Blend	<ul> <li>Select Color Blend Mode</li> <li>Select OK</li> <li>Select 3 different regions to blend</li> <li>Select OK</li> <li>Color will be assigned based on the number of the regions satisfied by the event</li> </ul>
Copy and Paste	
Copy Image	<ul> <li>Select a plot</li> <li>Right click and select Copy Plot Image</li> </ul>
Paste Image	<ul> <li>Open the desired program (e.g. PowerPoint, Word)</li> <li>Select desired area</li> <li>Select Edit &gt;&gt; Paste</li> </ul>
Copy Data	<ul> <li>Select plot</li> <li>Right click and select Copy Plot Data</li> </ul>
Paste Data	<ul> <li>Open the desired program (e.g. MS Excel)</li> <li>Select desired area</li> <li>Select Edit &gt;&gt; Paste</li> </ul>
Freeze Single Histogram Plots	<ul><li>Run a sample or retrieve a listmode file</li><li>Choose a histogram plot to apply freeze frame</li></ul>

- Move the cursor to the lower right corner of the plot Until >>>> appears
- Left click and select the Histogram tab
- Select Freeze Frame and then **OK**
- Run another sample or retrieve another file, an outline of the previous file appears on top of the new file
   You can repeat the process to achieve 2 freeze

frames on top of a new file

#### Select File >> Workspace Preferences

- Select Plot Display tab
- Select Baseline Offset On
- System adds a randomized positive Gaussian offset to thelow channels bringing the negative population out from channel zero on log displays

#### Listmode Compensation

Baseline Offset

- Select Listmode tab on the Resource Explorer
- Select desired listmode file
- Drag and drop it to the workspace
- Select Analysis >> LMD Quick COMP
- Adjust the slider bars to change the compensation
- The data responds accordingly
- Save recompensated file (file extension will be CMP)

# System Setup

Administration Functions	<ul> <li>Select a the user with administration privileges</li> <li>Enter the user password</li> <li>Select Admin button</li> <li>Select the user, enter both the user and administration passwords and select OK</li> </ul>
Add a User	<ul> <li>Select Add User</li> <li>Enter user name, first name, last name and title</li> <li>Select desired privileges</li> <li>Select Change and enter a user password</li> <li>Select OK</li> <li>Optional: Select Paths tab and change user paths</li> <li>Select OK</li> </ul>
Group Users	<ul> <li>Select Workgroups</li> <li>Select Edit and then Add</li> <li>Rename your workgroup as desired</li> <li>Select OK</li> <li>Select all users to be included in the workgroup</li> <li>Select OK</li> </ul>
Track User Usage	<ul> <li>Select Export Log</li> <li>Enter range of dates and select Next</li> <li>Select users to be included in the billing file</li> <li>Select Next</li> <li>Select to send to clipboard or file and select Delimiter</li> <li>Select Finish and then Close</li> </ul>
Unlock a User	<ul> <li>Select user</li> <li>Select Modify User</li> <li>Select Unlock User</li> </ul>
Archive	<ul> <li>Select Archive Data</li> <li>Select Archive location</li> <li>Select dates</li> <li>Select data to archive</li> <li>Select Archive and then OK</li> <li>Select Close</li> </ul>

Workspace Preferences	<ul> <li>Select File &gt;&gt; Workspace Preferences</li> </ul>
File Naming	<ul> <li>Select LMD Filename tab</li> <li>Select items to be part of the filename</li> <li>Select OK</li> </ul>
Plot Display	<ul> <li>Select Plot Display tab</li> <li>Select desired options</li> <li>Select OK</li> </ul>
Gating	<ul> <li>Select Gating tab</li> <li>Select desired options</li> <li>Select OK</li> </ul>
Acquisition	<ul> <li>Select Acquisition Options tab</li> <li>Select desired output options</li> <li>Select OK</li> </ul>
User Info	<ul> <li>Select User Info tab</li> <li>Enter desired user information</li> <li>Select OK</li> </ul>
Publish	<ul> <li>Select Publish tab</li> <li>Select desired publishing format options</li> <li>Select OK</li> </ul>