

AMPLIFICATION CFX Manager™ Software Data Analysis Quick Guide

Quantification Tab Data Viewing

The amplification chart displays traces of the relative fluorescence collected from each well at every cycle of the run.

Choose the fluorophore data you want to display by clicking the fluorophore checkboxes located under the amplification chart (Figure 1).

Click wells, rows, or columns in the well selector (Figure 1) to display fluorescence traces and data for selected wells. Selected wells are blue; unselected wells are light gray.

Position the cursor over a fluorescence trace, over a well in the well selector (Figure 1, well A3), over a cell in the data spreadsheet, or over a point on the standard curve to highlight corresponding information in all four locations.

Data Analysis Options

The software uses two modes for quantification cycle (C_q) determination: regression and single threshold. Select **Settings** from the menu bar, and then select **C_q Determination Mode** to change the determination method. In the Single Threshold mode, click and drag the threshold line to manually position the line.

CFX Manager software automatically subtracts the baseline from well data. Select **Settings** from the menu bar, and then select **Baseline Threshold** to change the baseline range for any well.

Click the **Plate Setup** button to choose:

- **View/Edit Plate** — edit well contents, create well groups, and add, remove, or exclude wells from analysis
- **Replace Plate File** — change plate layout to that of a previously saved plate file (.pltd)
- **Apply PrimePCR File** — change plate layout to that of a PrimePCR™ run file (.csv)

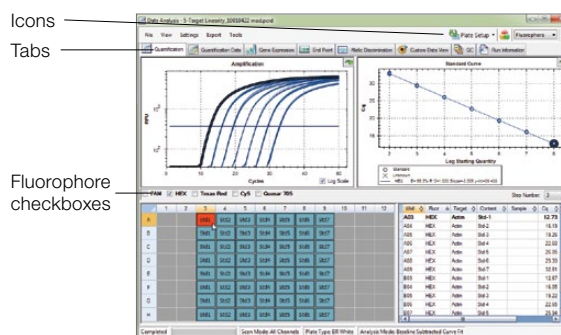


Fig. 1. Quantification tab in the Data Analysis window shows HEX data for all wells; information for well A3 is highlighted. Tab includes (counterclockwise from top left) the amplification chart, well selector, data spreadsheet, and standard curve.

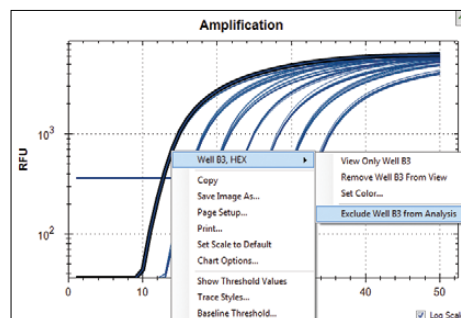


Fig. 2. Pop-up menu revealed by right clicking the amplification chart.

Chart Options

Right click any chart to copy or save the image, or to select **Chart Options** to modify the chart (Figure 2).

Select **Trace Styles** from the Settings menu or right click any chart to customize the trace colors for specific wells.

Left click any chart, then hold down and drag the cursor to zoom in.

Well Group Data Viewing

Use the Select Well Group dropdown menu on the toolbar to view data for a specific well group (Figure 3). The software treats well groups as independent runs, and each can be analyzed with its own settings and, if applicable, with its own standard curve.

Click the **Manage Well Groups** icon on the toolbar to add or modify well groups.

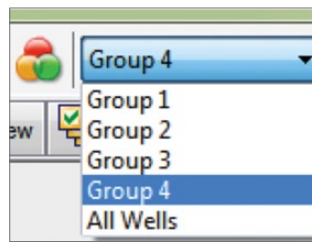


Fig. 3. Well group selection.

Quantification Data Tab

Display Options

Click the **Quantification Data** tab in the Data Analysis window to view numerical data calculations and statistics, including C_q, for each well and the mean and standard deviation for replicates (Figure 4).

Click any column header to sort rows based on that column.

Right click the spreadsheet and select **Sort** to sort rows based on multiple columns. Choose **Select Columns** to specify which columns will be displayed.

Left click any column header and drag the column to the left or to the right to rearrange the order of the columns in the spreadsheet.

Right click the spreadsheet and select to export data as a text, XML, Microsoft Excel, or HTML file (Figure 4).

Select **Plate** from the dropdown menu under the Quantification Data tab to display well data for a selected fluorophore in a plate grid format.

Creating a Report

Display Options

Select **Tools > Reports** from the toolbar menu of the Data Analysis window (Figure 1) to open the Report window (Figure 5).

Click checkboxes in the report options list to include specific information in the report, which is displayed in the preview window (Figure 5).

The ordering of categories and items within a report can be changed by clicking and dragging these to the desired relative position.

Click **File** on the toolbar and select **Save** to save the report as a PDF or select **Print** to print the report.

Save the report template for use with other data files if desired.

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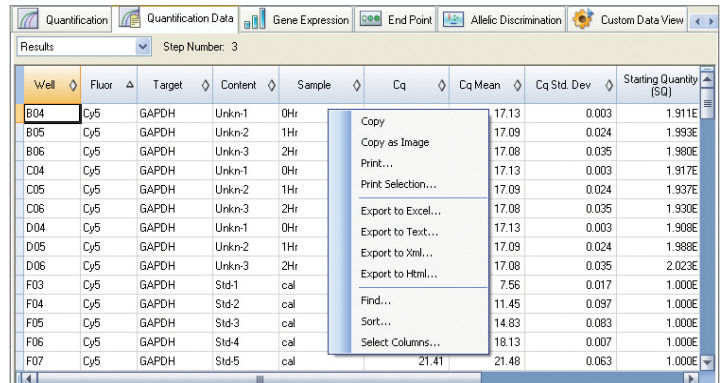


Fig. 4. Quantification data tab. Pop-up menu is revealed by right clicking the spreadsheet.

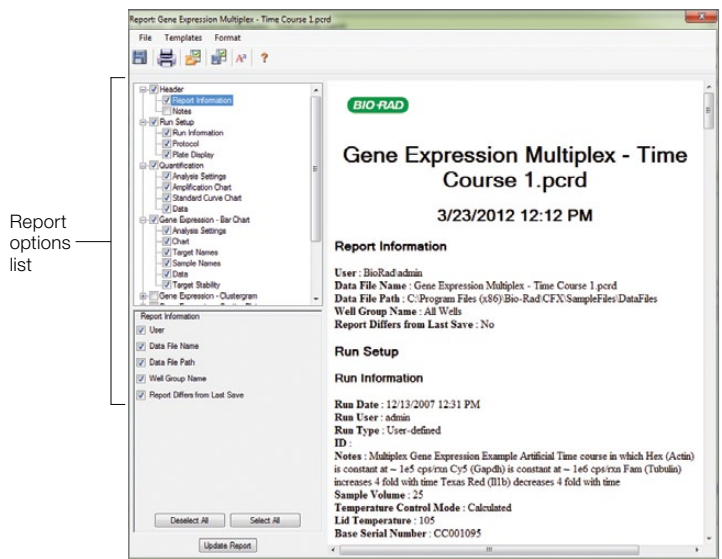


Fig. 5. Report window. Report options list includes checkboxes for choosing and ordering information to include in the report.



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