

## Technical Note

### LightCycler® 1536 Real-Time PCR System

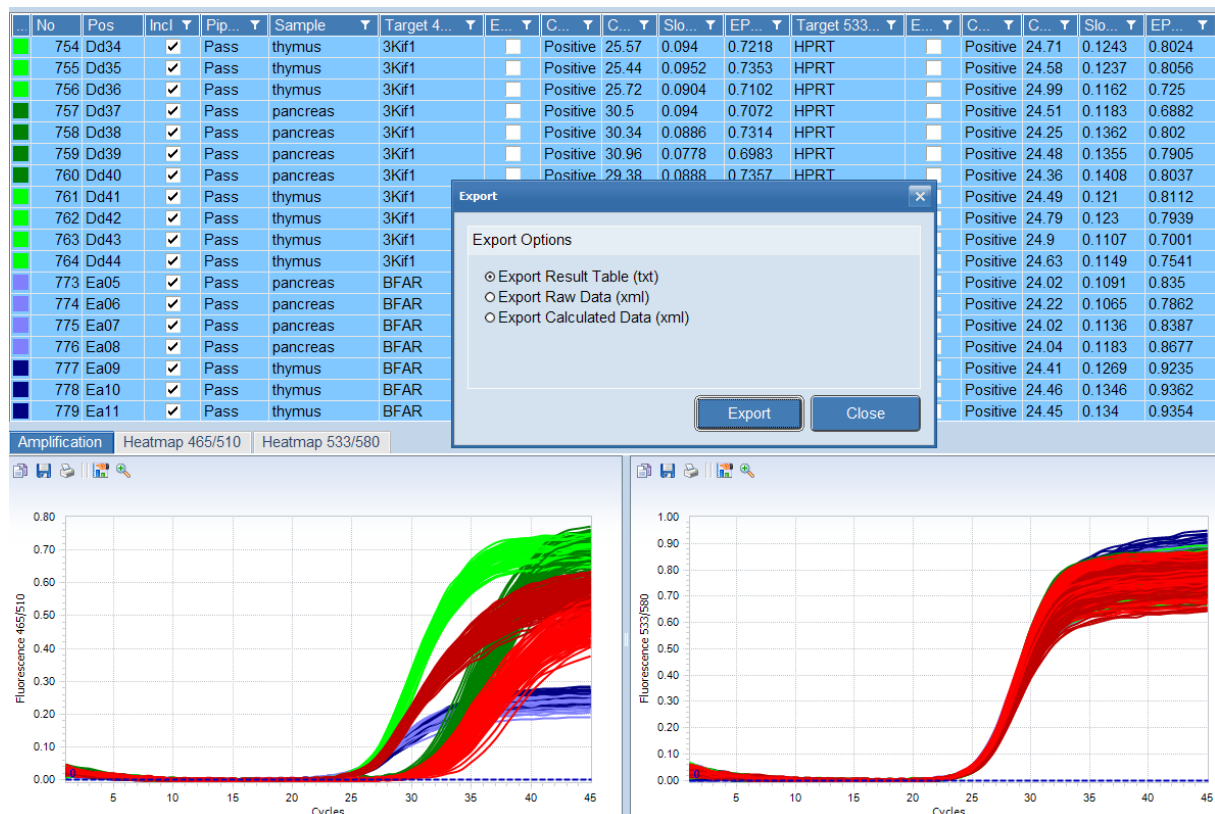
#### *Transferring Data from LightCycler® 1536 Software to qbase<sup>PLUS</sup> Software*

This Technical Note describes how to transfer data from LightCycler® 1536 software 1.0 to qbase<sup>PLUS</sup> software versions 1.4 and later (<http://www.qbaseplus.com>). qbase<sup>PLUS</sup> software uses sample and target (gene) names defined in LightCycler® 1536 Software and Cp (Cq) values calculated by LightCycler® 1536 Software for subsequent relative quantification analysis.

Both mono color and dual color LightCycler® 1536 experiments can be imported into qbase<sup>PLUS</sup> software.

### Exporting Data from LightCycler® 1536 Software

Export and save the Result Table txt file from a LightCycler® 1536 run to a dedicated folder.



#### Note:

For the subsequent relative quantification analysis in qbase<sup>PLUS</sup> software each position in the sample list must contain both a sample name and a target (gene) name.

## qbase<sup>PLUS</sup> Software

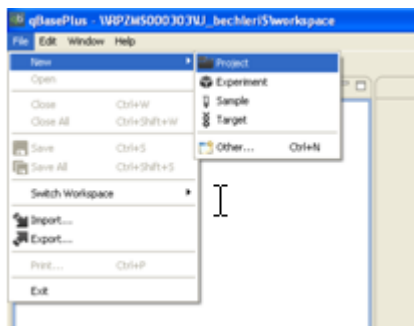
qbase<sup>PLUS</sup> software uses a hierarchical structure for data management in the following order:  
Project → Experiment → Run

Multiple qPCR runs can be merged within one experiment. Selected analysis settings are applied to all runs contained in an experiment.

Multiple experiments can be organized in one project.

## Importing Data into qbase<sup>PLUS</sup> Software

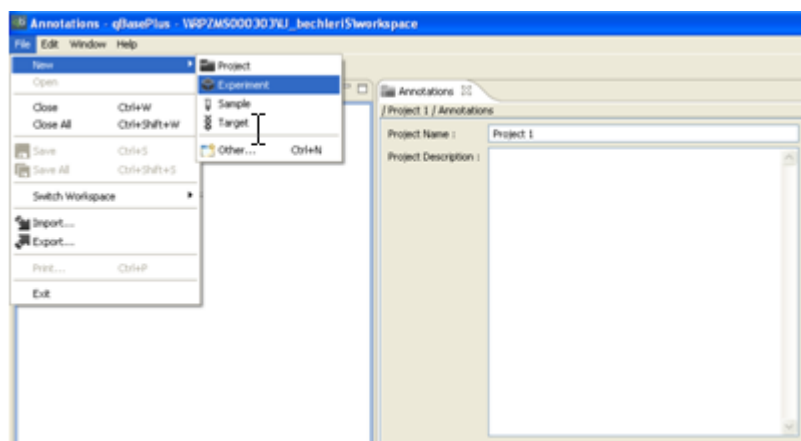
1. Open the qbase<sup>PLUS</sup> software.
2. Create a new project folder by selecting File – New – Project.  
Click the “Finish” button in the “New Project” window.  
A new project folder, named “Project 1”, is shown in the qbase<sup>PLUS</sup> explorer pane.



3. Create a new experiment folder by selecting File – New – Experiment.  
Select “Project 1” and click the “Finish” button in the “New Experiment” window.  
A new experiment subfolder, named “Experiment 1”, is stored under Project 1 – Experiments.

**or:**

Right Mouse click on “Experiments” in the “Projects 1” folder, then select “New Experiment”.



4. Start the import wizard by using one of the following modes:

- Select File – Import in the main menu.  
Select “Import Run” and click “Next”.

**or:**

- Click the “Import” button in the command bar.



Select “Import Run” and click “Next”.

**or:**

- Right Mouse click on “Runs” subfolder under “Experiment 1”.  
Select “Import Runs” from the context menu.

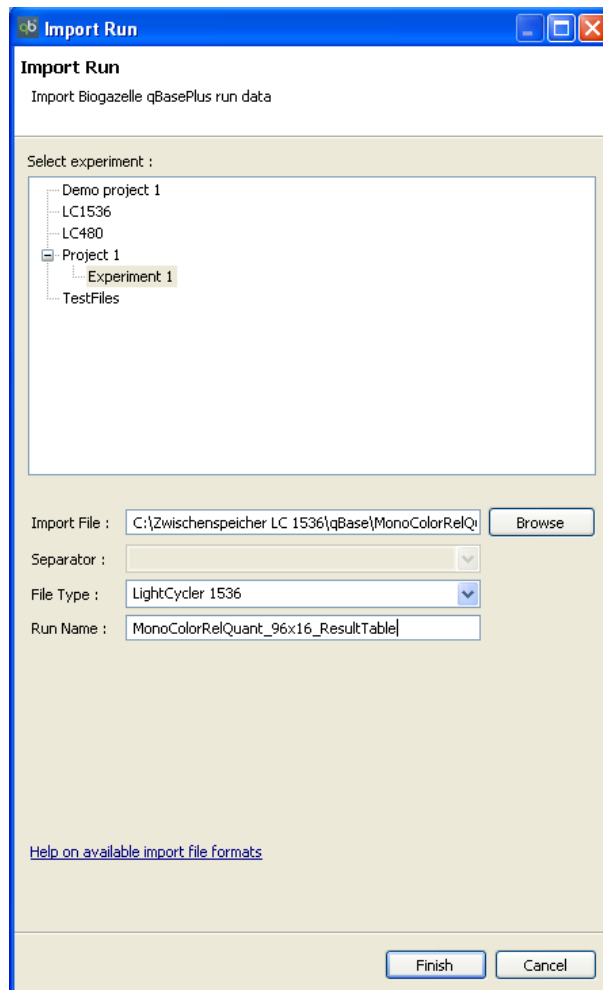
Select “Experiment 1” in the top part of the “Import Run” window for import of the run file(s).

Browse for the file(s) to be imported by clicking the “Browse” button and select txt file(s) from the LightCycler® 1536 software.

It is possible to import multiple txt files simultaneously using the Shift or Ctrl key.

Select “LightCycler 1536” in the File Type drop down menu.

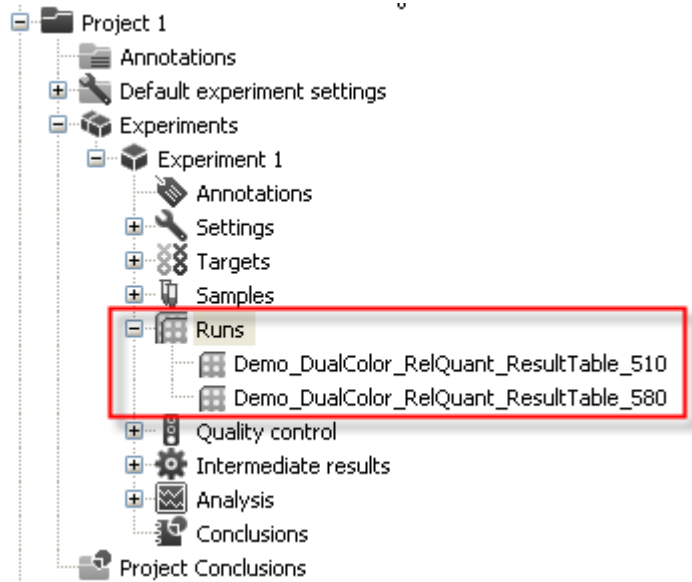
Click the “Finish” button to complete the import process and to close the run import wizard.



The imported runs are displayed under the “Runs” subfolder of “Experiment 1”.

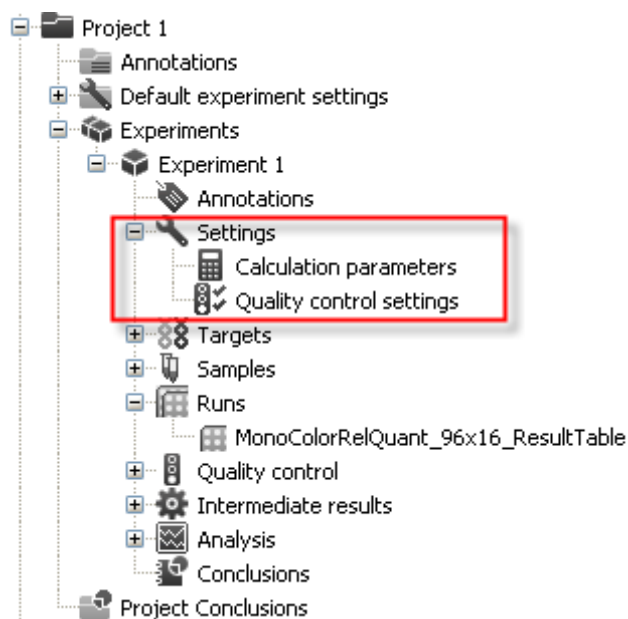
**Note:**

When importing a LightCycler® 1536 Dual Color experiment, the data of each channel are displayed as two separate run files in the qbase<sup>PLUS</sup> Explorer.

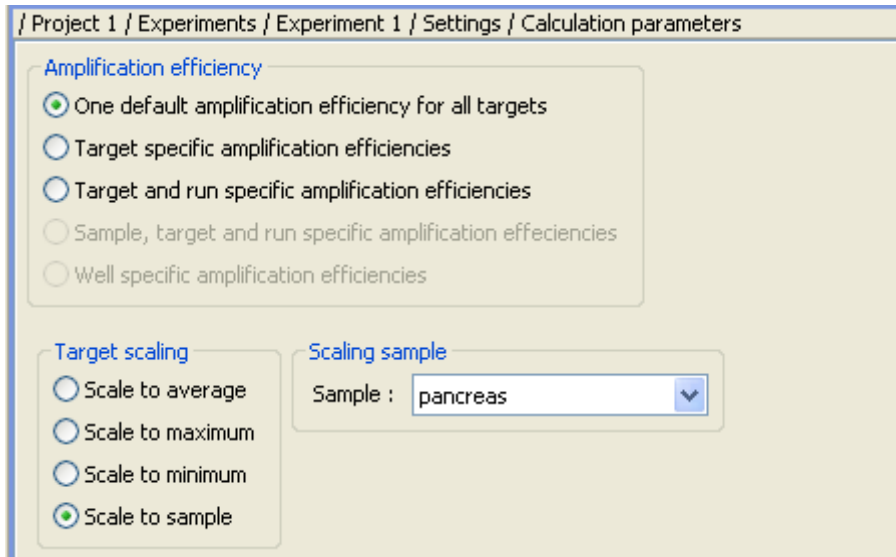


### Relative quantification analysis in qbase<sup>PLUS</sup> Software

- Define the relevant calculation settings by expanding the “Settings” folder under Experiment 1”.



By double clicking on “Calculation parameters” a new tab is opened in the main software pane.

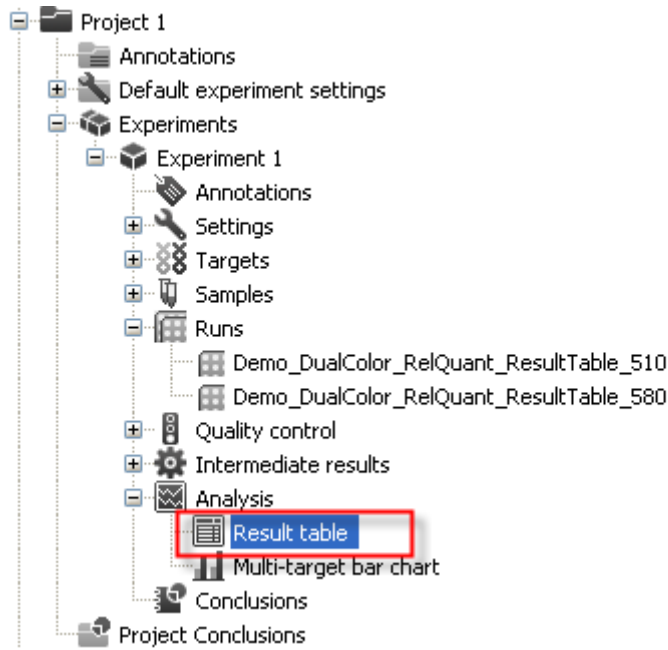


For Target scaling select for example “Scale to sample” and select a calibrator sample in the Sample drop down menu. This calibrator sample will get a relative quantity of 1.

- Under “Experiment 1” expand the Targets folder and then the “Targets of interests” folder.  
By default qbase<sup>PLUS</sup> lists all targets contained in the experiment as “Targets of interest”. Define at least one target as reference target to calculate normalized relative gene expression results. With right mouse click on the name of the reference target, select “Target Type – Reference” from the context menu.



- To view the results for a single target, double-click on the target in the project explorer pane.
- To view the results table, expand the “Analysis” folder under “Experiment 1” and double click on “Result Table”. This table can be copied or exported.

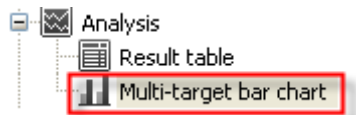


The calculated gene expression results are shown in a new tab in the main software pane.

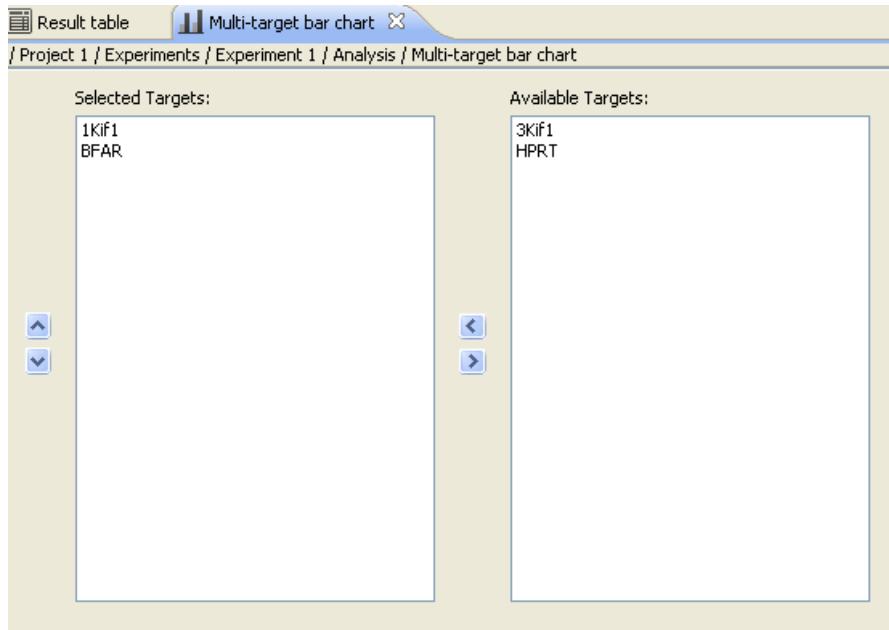
/ Project 1 / Experiments / Experiment 1 / Analysis / Result table

	HPRT	3Kif1	BFAR	1Kif1
pancreas	1.0000E0 ± 1.019E-2	1.000E0 ± 4.665E-2	1.000E0 ± 1.596E-2	1.000E0 ± 5.211E-2
thymus	1.0000E0 ± 1.387E-2	3.821E1 ± 5.942E-1	8.201E-1 ± 1.410E-2	5.363E1 ± 9.564E-1

- To display the bar chart double click on “Multi-target bar chart”.



Move the targets of interest from the “Available Targets” list into the “Selected Targets” list.



Click the “Chart” button in the bottom left corner in the “Multi-target bar chart” pane to view graphical results.

