LC480_to_RDES conversion

LC480 to RDES conversion is a program for the conversion of the raw fluorescence output of the LightCycler 480 into the RDES input format that is needed to create an RDML file for use in RDML-Tools (https://www.gear-genomics.com/rdml-tools/ described in: *Untergasser et al., (2021) Web-based LinRegPCR: application for the visualization and analysis of (RT)-qPCR amplification and melting data. BMC Bioinformatics 22: 398. doi: 10.1186/s12859-021-04306-1*). The RDML-tools enable you to perform the complete qPCR data analysis from raw fluorescence data to between-group statistics and bar graphs.

This special conversion program is needed because the LC480 exports one line per amplification cycle and melting curve temperature for every well on the plate which can lead to as much as 75000 lines in the LC480 txt output file. This program converts this text file into two row-by-column tables with amplification and melting data, respectively.

Before starting the LC480 to RDES conversion program you have to start Excel, with an empty book, to be able to export and save the fluorescence data in Excel. After opening the LC480 to RDES Conversion program you specify the LC480 txt output file from which the data need to be extracted and converted. The program identifies how many cycles you have run, extracts the cycle information per used well and converts the data into a table with a one-row-per-sample, one-column-per-cycle format. Similarly, the program extracts the melting curve data and converts them to a table with fluorescence values per temperature per reaction. Note that in both tables also all empty wells are extract. They are needed in the RDML-Tools and should NOT be removed.

Before exporting both tables to Excel you have to indicate the experiment name, say ‘expX’, which is used in the name of the two export sheets, named expX_Amplif and expX_Melt, respectively. The experiment name that you give should not be more than 12 characters.

The output in Excel also contains 6 columns with only a fixed header row. The first five columns, describing Sample, Sample Type, Target, Target Type and Dye have to be completed in Excel. The 6th column indicated Cq or Tm can stay empty. The required entries for Sample Type and Target Type are described in detail in the manuscript: *Untergasser et al., (2023) Disclosing quantitative RT-PCR raw data during manuscript submission: a call for action. Mol Oncol. 17: 713-717. doi: 10.1002/1878-0261.13418.* Samples with the same name are treated as technical replicate reactions. For empty wells, these columns are not filled. The contents of these columns should be the same in the expX_Amplif and expX_Melt sheets.

Moreover, when the program found that in the text file that you got from the LightCycler uses the comma as decimal separator it will change this to the decimal point, required by RDML.