Luminex Xponent: Plate-level Quality Control

Sahal Thahir & Jeff Bailey

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Contents

Aim

This code aims to perform plate-level quality control analysis for Luminex studies through bead counts, Background MFI, and Standard curve analysis.

Instructions

1. **Modify YAML Parameters:**
   - Open this document in RStudio.
   - Edit the YAML parameters at the top of the document:
     - `input_file`: Path to your input CSV file.
     - `min_beadcount`: Set the minimum bead count threshold.
     - `min_Rsquared`: Set the minimum R-squared value for standard curve QC.
     - `file_path`: Specify the output directory for plots and PDFs.
     - '
2. **Run the Analysis:**
   - After modifying the YAML parameters, click the *knit* button at the top of the page to generate the report.

Code Explanation

Dependencies

- R version 4.3.1 (2023-06-16)
  - `tidyverse` (v. 2.0.0)
  - `here` (v. 1.0.1)
  - `gridExtra` (v. 2.3)

This code chunk will ensure these packages are installed and loaded.

Import and Data Extraction

1. **Raw data import:** Import raw data CSV file into R.

```r
# No input required
  #YAML extraction
    # Read parameters from YAML header
      input_file <- params$input_file
      min_beadcount <- params$min_beadcount
      min_Rsquared <- params$min_Rsquared
      file_path <- params$file_path
```

```
        background_col <- params$background

  # Raw data import
        lines <- readLines(input_file)
```

Warning in readLines(input_file): incomplete final line found on
'data/raw/luminex/Control Run 13 June 2024_20240613_161509.csv'

2. **Extract dataframes:** find median MFI and bead count data. This includes a check to ensure the median MFI and beadcount dataframes align.

3. **Identify low bead count wells:** this searches the beadcount data frame for well-antigen data with low bead count, as set in the YAML.This will export a csv file of the samples which will need to be rerun.

The bead QC list has been exported to data/qc/Control Run 13 June 2024_20240613_161509_beadqc_low_df

**Visualization of bead count data**

This code generates a well schematic identifying wells identified as having low bead counts. as well as a heatmap of antigen/well locations and the quantity of beads, to assist in determining the cause of low bead counts (low antigen concentration in mastermix, vortexing between wells, etc.)

96−well schematic of Plate: Control Run 13 June 2024_20240613_161509
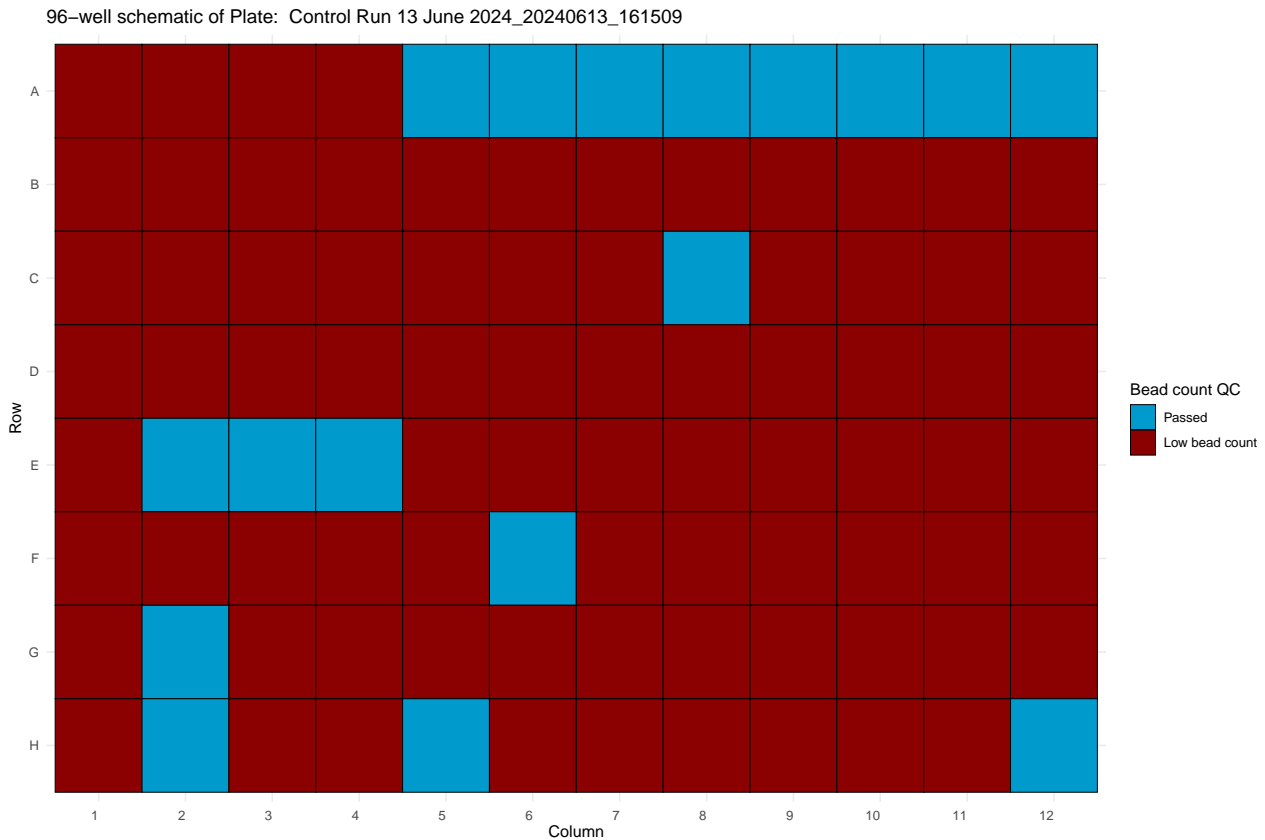
```
## # A tibble: 772 x 4
##    Location Sample       Antigen      Plate
##    <chr>    <chr>        <chr>        <chr>
## 1 1(1,A1)  Background0 BSA          Control Run 13 June 2024_20240613_161509
## 2 1(1,A1)  Background0 Rickettsia   Control Run 13 June 2024_20240613_161509
## 3 1(1,A1)  Background0 Pf.MSP1      Control Run 13 June 2024_20240613_161509
```

```
##  4 1(1,A1)  Background0 Dengue.3    Control Run 13 June 2024_20240613_161509
##  5 1(1,A1)  Background0 Dengue.4    Control Run 13 June 2024_20240613_161509
##  6 1(1,A1)  Background0 CCHFV       Control Run 13 June 2024_20240613_161509
##  7 1(1,A1)  Background0 Dengue.2    Control Run 13 June 2024_20240613_161509
##  8 1(1,A1)  Background0 Measles     Control Run 13 June 2024_20240613_161509
##  9 1(1,A1)  Background0 Chikungunya Control Run 13 June 2024_20240613_161509
## 10 1(1,A1)  Background0 Pv.MSP1     Control Run 13 June 2024_20240613_161509
## # i 762 more rows
```

```
## The bead QC list has been exported to data/qc/Control Run 13 June 2024_20240613_161509_beadqc_low_df
```

```
## The bead QC report has been exported to data/qc/Control Run 13 June 2024_20240613_161509_beadqc.png
```


Quality Control: Bead count for each antigen Control Run 13 June 2024_20240613_161509
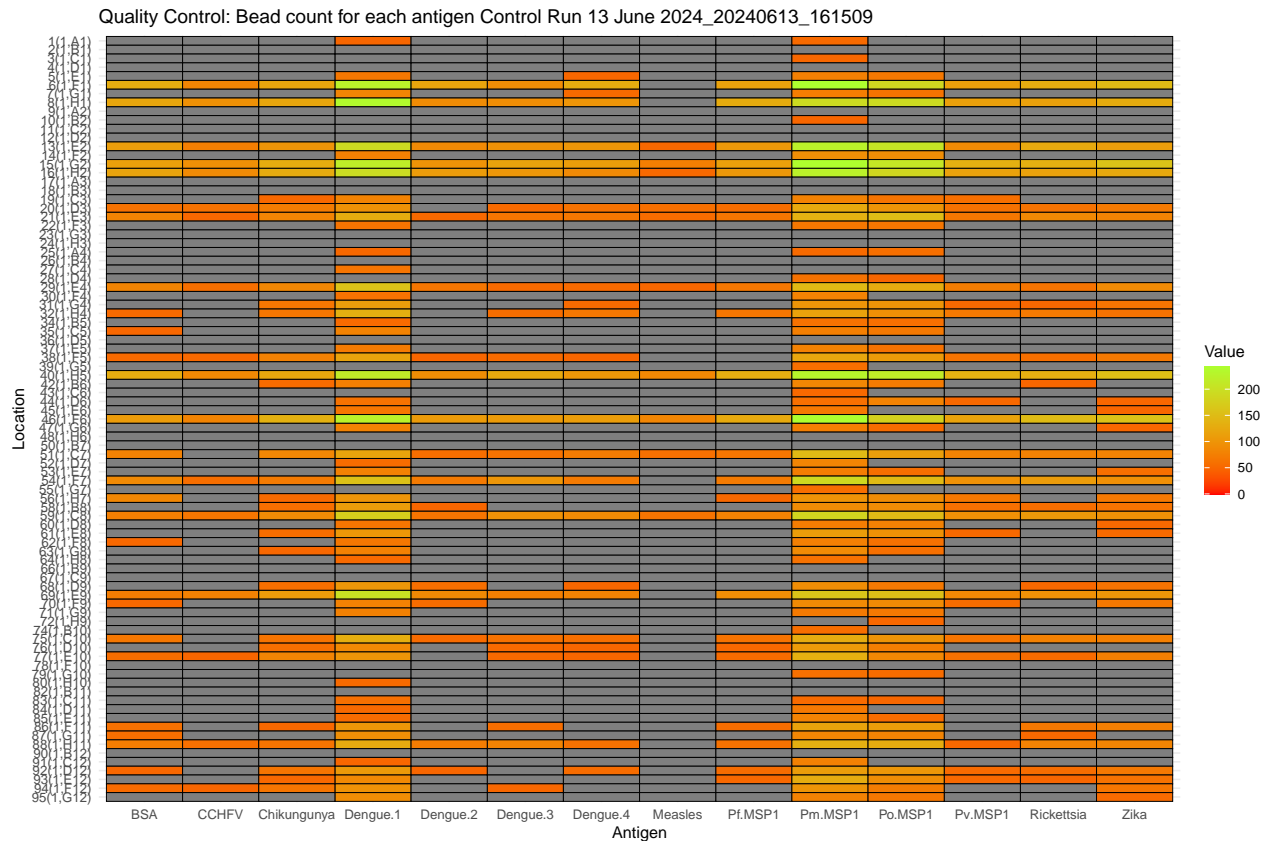
4. **Modify Median MFI data:** All plate locations with a low beadcount with be switched to a `NA` value for standard curve evaluation

```
## Median MFI data has been modified for low bead count wells.
```

5. **Evaluate standard curves:** Make sure the background and standard analytes are defined in the YAML params

This first chunk conducts background MFI subtraction, based on the analyte defined in the YAML.

```
## Background MFI subtraction completed.
```

This code chunk

```
## No valid plots to display.
```

```
## [1] Analyte   R_squared Equation
## <0 rows> (or 0-length row.names)
```

Create quality controlled Median MFI data

```
## [1] Analyte   R_squared Equation   min_Rsq
## <0 rows> (or 0-length row.names)
```

```
## [1] "This plate has failed plate-level QC for standard curves."
## [1] "No analytes met the preset minimum R-squared (see min_Rsquared in YAML)."
## [1] "All MFI values from this plate will display as 'Failed Plate QC'."
```

```
## Plate failed QC. All median MFI data set to 'Failed QC (standards)'.
```

```
## The cleaned median MFI data has been exported to data/clean/luminex/Control Run 13 June 2024_2024061
```

Developer notes

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