



### **Document information**

Software: realplex 2.2

File Name: EPPENDORF\Lorenzo\QPCR18.06.2

Printed by: EPPENDORF
Created: Jun/18/2018 17:23

Serial No. Thermo Module: 6325 30387 Serial No. realplex Module.: 630011465

Acquisition Start Time: EPPENDORF Jun/18/2018 17:26
Acquisition End Time: EPPENDORF Jun/18/2018 18:54
Last updated: EPPENDORF Apr/06/2018 12:28

Background: Sarstedt-20µl Sep/12/2011 10:28 Color Calibration: SYBR Mar/12/2018 15:31

QPCR18.06.2018 Quantification Jun/18/2018 18:54

Inverted Data: OFF

Comment:

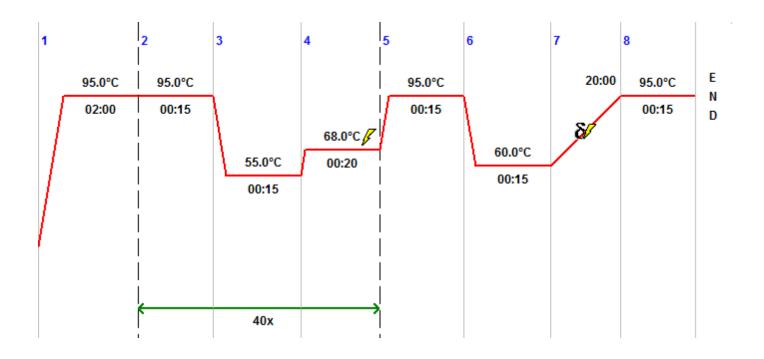


# **Plate layout**

|   | 1       | 2       | 3       | 4       | 5       | 6       | 7       | 8       | 9       | 10      | 11      | 12      |
|---|---------|---------|---------|---------|---------|---------|---------|---------|---------|---------|---------|---------|
| Α | ILWE_A  |
|   | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 |
| В | ILWE_A  |
|   | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 |
| С | ILWE_A  |
|   | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 |
| D | ILWE_A  |
|   | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 |
| E | ILWE_A  |
|   | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 |
| F | ILWE_A  |
|   | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 |
| G | CEWE    | CEWE    | CEWE    | CEWE    | CEWE    | CEWE    | ILWE_A  | ILWE_A  | ILWE_A  | ILWE_A  | ILWE_A  | ILWE_A  |
|   | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 | 1: 1.00 |
| Н | NTC     | NTC     | NTC     | NTC     | NTC     | NTC     | water   | water   | water   | water   | water   | water   |



# **PCR Program**



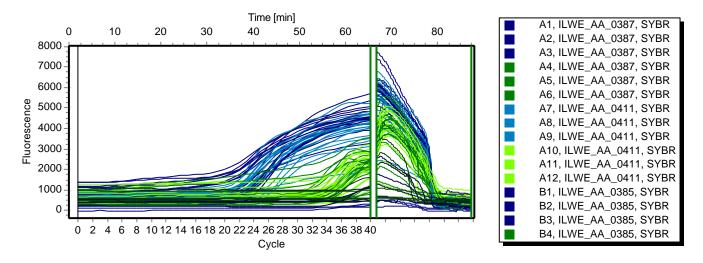
## **Program Header**

| Lid Temp   | 105 °C   | TSP Heated Lid                   | Yes |
|------------|----------|----------------------------------|-----|
| Temp. Mode | Standard | Switch off lid at low block temp | No  |
| Impulse    | No       | Simulate Mastercycler gradient   | No  |

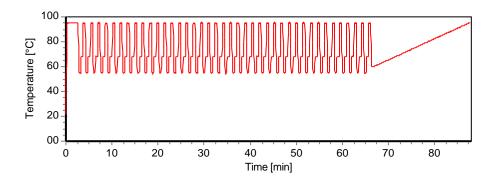


### **Raw Data SYBR**

#### Fluorescence Profile



## **Temperature Profile**





## **Quantification SYBR**

| Pos             | Name         | Ct SYBR | Ct Mean<br>SYBR | Ct Dev.<br>SYBR | Amount<br>SYBR<br>[Copies] | Amount<br>Mean SYBR | Amount<br>Dev. SYBR | Target SYBR |
|-----------------|--------------|---------|-----------------|-----------------|----------------------------|---------------------|---------------------|-------------|
| ! <b>■</b> A1   | ILWE_AA_0387 | 18.86   | 18.60           | 0.40            | 1.00                       |                     |                     | mouse       |
| • ■ A2          | ILWE_AA_0387 | 18.14   | 18.60           | 0.40            | 1.00                       |                     |                     | mouse       |
| ! <b>■</b> A3   | ILWE_AA_0387 | 18.80   | 18.60           | 0.40            | 1.00                       |                     |                     | mouse       |
| ! <b></b> ■ A4  | ILWE_AA_0387 | 34.91   | 33.79           | 0.97            | 1.00                       |                     |                     | eimeria     |
| ! <b></b> ■ A5  | ILWE_AA_0387 | 33.22   | 33.79           | 0.97            | 1.00                       |                     |                     | eimeria     |
| ! <b></b> ■ A6  | ILWE_AA_0387 | 33.24   | 33.79           | 0.97            | 1.00                       |                     |                     | eimeria     |
| ! <b> </b>      | ILWE_AA_0411 | 23.32   | 23.44           | 0.14            | 1.00                       |                     |                     | mouse       |
| ! <b></b> ■ A8  | ILWE_AA_0411 | 23.42   | 23.44           | 0.14            | 1.00                       |                     |                     | mouse       |
| ! <b> </b> ■ A9 | ILWE_AA_0411 | 23.60   | 23.44           | 0.14            | 1.00                       |                     |                     | mouse       |
| <b>!</b>        | ILWE_AA_0411 | 33.31   | 33.82           | 0.56            | 1.00                       |                     |                     | eimeria     |
| ! <b> </b>      | ILWE_AA_0411 | 33.72   | 33.82           | 0.56            | 1.00                       |                     |                     | eimeria     |
| ! <b> </b>      | ILWE_AA_0411 | 34.42   | 33.82           | 0.56            | 1.00                       |                     |                     | eimeria     |
| <b>!</b> ■ B1   | ILWE_AA_0385 | 21.73   | 21.63           | 0.32            | 1.00                       |                     |                     | mouse       |
| <b>!</b> ■ B2   | ILWE_AA_0385 | 21.89   | 21.63           | 0.32            | 1.00                       |                     |                     | mouse       |
| <b>!</b> ■ B3   | ILWE_AA_0385 | 21.28   | 21.63           | 0.32            | 1.00                       |                     |                     | mouse       |
| <b>!</b> ■ B4   | ILWE_AA_0385 | 34.23   | 34.03           | 0.59            | 1.00                       |                     |                     | eimeria     |
| ! <b>■</b> B5   | ILWE_AA_0385 | 34.49   | 34.03           | 0.59            | 1.00                       |                     |                     | eimeria     |
| <b>!</b> ■ B6   | ILWE_AA_0385 | 33.36   | 34.03           | 0.59            | 1.00                       |                     |                     | eimeria     |
| <b>!</b> ■ B7   | ILWE_AA_0511 | 22.53   | 22.60           | 0.13            | 1.00                       |                     |                     | mouse       |
| ! <b>■</b> B8   | ILWE_AA_0511 | 22.74   | 22.60           | 0.13            | 1.00                       |                     |                     | mouse       |
| ! <b>■</b> B9   | ILWE_AA_0511 | 22.52   | 22.60           | 0.13            | 1.00                       |                     |                     | mouse       |
| <b>!</b> ■ B10  | ILWE_AA_0511 | 34.13   | 35.54           | 1.27            | 1.00                       |                     |                     | eimeria     |
| ! <b>∏</b> □B11 | ILWE_AA_0511 | 35.92   | 35.54           | 1.27            | 1.00                       |                     |                     | eimeria     |
| <b>!</b> ■ B12  | ILWE_AA_0511 | 36.57   | 35.54           | 1.27            | 1.00                       |                     |                     | eimeria     |
| ! <b>■</b> C1   | ILWE_AA_0384 | 22.79   | 22.86           | 0.25            | 1.00                       |                     |                     | mouse       |
| ! <b>■</b> C2   | ILWE_AA_0384 | 23.13   | 22.86           | 0.25            | 1.00                       |                     |                     | mouse       |
| <b>i</b>        | ILWE_AA_0384 | 22.65   | 22.86           | 0.25            | 1.00                       |                     |                     | mouse       |
| ! <b>■</b> C4   | ILWE_AA_0384 | 31.88   | 33.24           | 1.25            | 1.00                       |                     |                     | eimeria     |
| ! <b>□</b> C5   | ILWE_AA_0384 | 34.34   | 33.24           | 1.25            | 1.00                       |                     |                     | eimeria     |
| <b>i</b>        | ILWE_AA_0384 | 33.50   | 33.24           | 1.25            | 1.00                       |                     |                     | eimeria     |
| <b>!</b>        | ILWE_AA_0512 | 20.71   | 21.44           | 0.82            | 1.00                       |                     |                     | mouse       |
| <b>i</b>        | ILWE_AA_0512 | 21.28   | 21.44           | 0.82            | 1.00                       |                     |                     | mouse       |
| <b>i</b>        | ILWE_AA_0512 | 22.32   | 21.44           | 0.82            | 1.00                       |                     |                     | mouse       |
| <b>!</b> ☐ C10  | ILWE_AA_0512 | 31.90   | 33.07           | 1.34            | 1.00                       |                     |                     | eimeria     |
| ! <b>∏</b>      | ILWE_AA_0512 | 32.78   | 33.07           | 1.34            | 1.00                       |                     |                     | eimeria     |



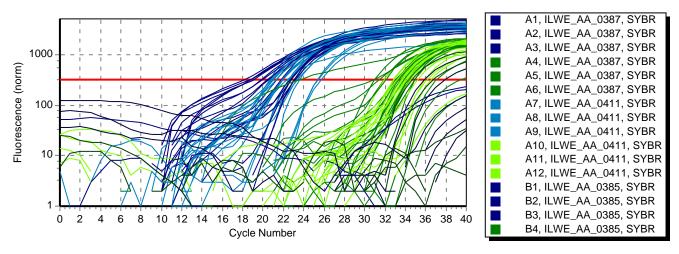
| Pos             | Name         | Ct SYBR | Ct Mean<br>SYBR | Ct Dev.<br>SYBR | Amount<br>SYBR<br>[Copies] | Amount<br>Mean SYBR | Amount<br>Dev. SYBR | Target SYBR |
|-----------------|--------------|---------|-----------------|-----------------|----------------------------|---------------------|---------------------|-------------|
| C12             | ILWE_AA_0512 | 34.53   | 33.07           | 1.34            | 1.00                       |                     |                     | eimeria     |
| . □ □ D1        | ILWE_AA_0388 | 19.06   | 19.39           | 0.91            | 1.00                       |                     |                     | mouse       |
| . D2            | ILWE_AA_0388 | 18.69   | 19.39           | 0.91            | 1.00                       |                     |                     | mouse       |
| ! <b>■</b> D3   | ILWE_AA_0388 | 20.42   | 19.39           | 0.91            | 1.00                       |                     |                     | mouse       |
| <b>!</b> ■ D4   | ILWE_AA_0388 | 32.03   | 32.01           | 1.02            | 1.00                       |                     |                     | eimeria     |
| ! <b>■</b> D5   | ILWE_AA_0388 | 33.02   | 32.01           | 1.02            | 1.00                       |                     |                     | eimeria     |
| <b>!</b> ■ D6   | ILWE_AA_0388 | 30.98   | 32.01           | 1.02            | 1.00                       |                     |                     | eimeria     |
| <b>!</b>        | ILWE_AA_0465 | 25.37   | 25.48           | 0.16            | 1.00                       |                     |                     | mouse       |
| <b>!</b> ■ D8   | ILWE_AA_0465 | 25.41   | 25.48           | 0.16            | 1.00                       |                     |                     | mouse       |
| <b>!</b> ■ D9   | ILWE_AA_0465 | 25.67   | 25.48           | 0.16            | 1.00                       |                     |                     | mouse       |
| <b>!</b> ☐ D10  | ILWE_AA_0465 | 37.88   | 36.06           | 2.56            | 1.00                       |                     |                     | eimeria     |
| ! <b>□</b> □D11 | ILWE_AA_0465 | 34.25   | 36.06           | 2.56            | 1.00                       |                     |                     | eimeria     |
| <b>!</b> ☐ D12  | ILWE_AA_0465 |         | 36.06           | 2.56            | 1.00                       |                     |                     | eimeria     |
| ! <b>∭</b> ■E1  | ILWE_AA_0389 |         |                 |                 | 1.00                       |                     |                     | mouse       |
| ! <b>■</b> E2   | ILWE_AA_0389 |         |                 |                 | 1.00                       |                     |                     | mouse       |
| ! <b></b> ■E3   | ILWE_AA_0389 |         |                 |                 | 1.00                       |                     |                     | mouse       |
| <b>!</b> ■ E4   | ILWE_AA_0389 |         | 38.29           | 1.65            | 1.00                       |                     |                     | eimeria     |
| ! <b>■</b> E5   | ILWE_AA_0389 | 39.45   | 38.29           | 1.65            | 1.00                       |                     |                     | eimeria     |
| <b>!</b> ■ E6   | ILWE_AA_0389 | 37.13   | 38.29           | 1.65            | 1.00                       |                     |                     | eimeria     |
| ! <b>■ E</b> 7  | ILWE_AA_0466 | 21.91   | 21.87           | 0.04            | 1.00                       |                     |                     | mouse       |
| ! <b></b> ■E8   | ILWE_AA_0466 | 21.83   | 21.87           | 0.04            | 1.00                       |                     |                     | mouse       |
| <b>!</b> ■ E9   | ILWE_AA_0466 | 21.87   | 21.87           | 0.04            | 1.00                       |                     |                     | mouse       |
| ! <b></b> ■E10  | ILWE_AA_0466 | 33.91   | 34.54           | 1.40            | 1.00                       |                     |                     | eimeria     |
| ! <b></b>       | ILWE_AA_0466 | 33.56   | 34.54           | 1.40            | 1.00                       |                     |                     | eimeria     |
| ! <b>∏</b>      | ILWE_AA_0466 | 36.15   | 34.54           | 1.40            | 1.00                       |                     |                     | eimeria     |
| ! <b></b>       | ILWE_AA_0288 | 23.04   | 22.95           | 0.16            | 1.00                       |                     |                     | mouse       |
| <b>!</b>        | ILWE_AA_0288 | 23.05   | 22.95           | 0.16            | 1.00                       |                     |                     | mouse       |
| <b>!</b>        | ILWE_AA_0288 | 22.77   | 22.95           | 0.16            | 1.00                       |                     |                     | mouse       |
| <b>!</b>        | ILWE_AA_0288 | 34.93   | 34.37           | 0.55            | 1.00                       |                     |                     | eimeria     |
| ! <b></b>       | ILWE_AA_0288 | 34.34   | 34.37           | 0.55            | 1.00                       |                     |                     | eimeria     |
| <b>!</b>        | ILWE_AA_0288 | 33.83   | 34.37           | 0.55            | 1.00                       |                     |                     | eimeria     |
| ! <b>∏ ■</b> F7 | ILWE_AA_0467 | 20.63   | 20.82           | 0.17            | 1.00                       |                     |                     | mouse       |
| ! <b></b>       | ILWE_AA_0467 | 20.91   | 20.82           | 0.17            | 1.00                       |                     |                     | mouse       |
| <b>!</b>        | ILWE_AA_0467 | 20.92   | 20.82           | 0.17            | 1.00                       |                     |                     | mouse       |
| ! <b>∏</b>      | ILWE_AA_0467 | 32.83   | 33.74           | 1.33            | 1.00                       |                     |                     | eimeria     |
| <b>!</b>        | ILWE_AA_0467 | 33.13   | 33.74           | 1.33            | 1.00                       |                     |                     | eimeria     |
| <b>!</b>        | ILWE_AA_0467 | 35.26   | 33.74           | 1.33            | 1.00                       |                     |                     | eimeria     |
| <b>!</b>        | CEWE_AA_0381 | 19.82   | 20.10           | 0.39            | 1.00                       |                     |                     | mouse       |
| ! <b>∏ G</b> 2  | CEWE_AA_0381 | 19.93   | 20.10           | 0.39            | 1.00                       |                     |                     | mouse       |



| Pos                   | Name         | Ct SYBR | Ct Mean<br>SYBR | Ct Dev.<br>SYBR | Amount<br>SYBR<br>[Copies] | Amount<br>Mean SYBR | Amount<br>Dev. SYBR | Target SYBR |
|-----------------------|--------------|---------|-----------------|-----------------|----------------------------|---------------------|---------------------|-------------|
| . G3                  | CEWE_AA_0381 | 20.55   | 20.10           | 0.39            | 1.00                       |                     |                     | mouse       |
| . <b>G</b> 4          | CEWE_AA_0381 | 23.49   | 28.63           | 4.46            | 1.00                       |                     |                     | eimeria     |
| G5                    | CEWE_AA_0381 | 30.95   | 28.63           | 4.46            | 1.00                       |                     |                     | eimeria     |
| . <b>G</b> 6          | CEWE_AA_0381 | 31.44   | 28.63           | 4.46            | 1.00                       |                     |                     | eimeria     |
| . <b>□ G</b> 7        | ILWE_AA_0468 | 20.84   | 21.03           | 0.20            | 1.00                       |                     |                     | mouse       |
| ! <b>■ G</b> 8        | ILWE_AA_0468 | 21.01   | 21.03           | 0.20            | 1.00                       |                     |                     | mouse       |
| . <b>□ G</b> 9        | ILWE_AA_0468 | 21.24   | 21.03           | 0.20            | 1.00                       |                     |                     | mouse       |
| G10                   | ILWE_AA_0468 | 32.89   | 33.53           | 0.57            | 1.00                       |                     |                     | eimeria     |
| G11                   | ILWE_AA_0468 | 33.74   | 33.53           | 0.57            | 1.00                       |                     |                     | eimeria     |
| G12                   | ILWE_AA_0468 | 33.97   | 33.53           | 0.57            | 1.00                       |                     |                     | eimeria     |
| - <mark>∏</mark> ■ H1 | NTC          | -       |                 |                 | -                          |                     |                     | mouse       |
| -TH2                  | NTC          | -       |                 |                 | -                          |                     |                     | mouse       |
| - <b>∐</b> ■H3        | NTC          | 35.89   |                 |                 | -                          |                     |                     | mouse       |
| <b>-</b> ■  H4        | NTC          | -       | 35.73           | 1.47            | -                          |                     |                     | eimeria     |
| - <b>∐</b> ■H5        | NTC          | 36.76   | 35.73           | 1.47            | -                          |                     |                     | eimeria     |
| <b>-</b> ☐ ■ H6       | NTC          | 34.69   | 35.73           | 1.47            | -                          |                     |                     | eimeria     |
| - <b>∐</b> ■H7        | water        | -       |                 |                 | -                          |                     |                     | mouse       |
| - <b>∐</b> ■H8        | water        | -       |                 |                 | -                          |                     |                     | mouse       |
| - <b>T</b> H9         | water        | 37.86   |                 |                 | -                          |                     |                     | mouse       |
| -<br>H10              | water        | -       |                 |                 | -                          |                     |                     | eimeria     |
| -T H11                | water        | -       |                 |                 | -                          |                     |                     | eimeria     |
| _<br>■H12             | water        | -       |                 |                 | -                          |                     |                     | eimeria     |



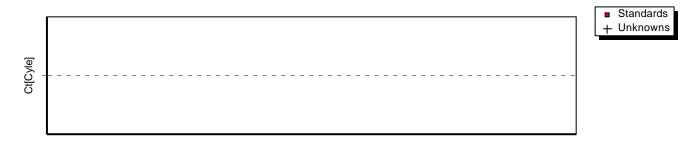
## **Amplification Plot**



Threshold 317 (Noiseband)

Baseline automatic, Drift correction OFF

#### Standard curve



Amount[Copies]

Slope - R^2 - Y-Intercept - Efficiency -