

Document information

Software: realplex 2.2

File Name: EPPENDORF\Svenja\ileumplate5

Printed by: EPPENDORF

Created: Nov/13/2018 09:55

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

Acquisition Start Time: EPPENDORF Nov/13/2018 09:59
Acquisition End Time: EPPENDORF Nov/13/2018 11:27
Last updated: EPPENDORF Nov/06/2018 18:40

Eppendorf

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

ileumplate5 Quantification Nov/13/2018 11:30

Melting Curve Nov/13/2018 11:30

Inverted Data: OFF

Comment:

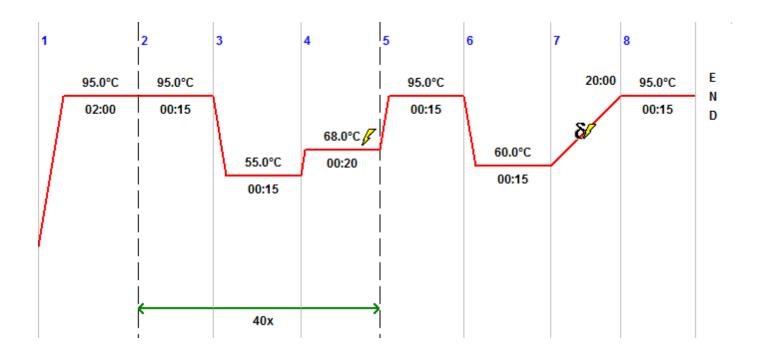


Plate layout

| | 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 | 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



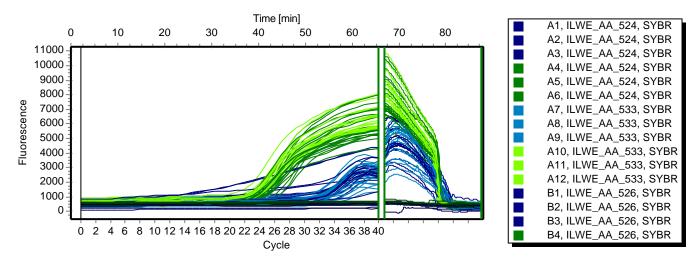
Program Header

| 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 |

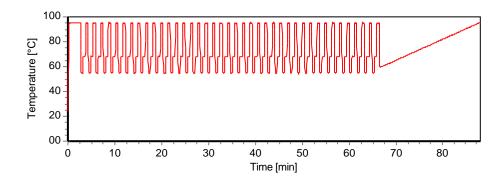


Raw Data SYBR

Fluorescence Profile



Temperature Profile





Quantification SYBR

| Pos | Name | Ct SYBR | Ct Mean SYBR | Ct Dev. SYBR | Amount SYBR [Copies] | Amount Mean SYBR | Amount Dev. SYBR | Target SYBR |
|----------------|-------------|---------|-----------------|-----------------|----------------------------|---------------------|---------------------|-------------|
| <u> </u> | ILWE_AA_524 | 32.53 | 32.25 | 0.25 | 1.00 | | | eimeria |
| <u>.</u> ■ A2 | ILWE_AA_524 | 32.18 | 32.25 | 0.25 | 1.00 | | | eimeria |
| . ■ A3 | ILWE_AA_524 | 32.04 | 32.25 | 0.25 | 1.00 | | | eimeria |
| ! ■ A4 | ILWE_AA_524 | 23.78 | 23.73 | 0.17 | 1.00 | | | mouse |
| . A5 | ILWE_AA_524 | 23.54 | 23.73 | 0.17 | 1.00 | | | mouse |
| | ILWE_AA_524 | 23.86 | 23.73 | 0.17 | 1.00 | | | mouse |
| ! ■ A7 | ILWE_AA_533 | 31.48 | 31.18 | 0.48 | 1.00 | | | eimeria |
| ! ■ A8 | ILWE_AA_533 | 30.63 | 31.18 | 0.48 | 1.00 | | | eimeria |
| ! ■ A9 | ILWE_AA_533 | 31.43 | 31.18 | 0.48 | 1.00 | | | eimeria |
| ! | ILWE_AA_533 | 21.50 | 21.35 | 0.26 | 1.00 | | | mouse |
| ! | ILWE_AA_533 | 21.49 | 21.35 | 0.26 | 1.00 | | | mouse |
| ! ■ A12 | ILWE_AA_533 | 21.05 | 21.35 | 0.26 | 1.00 | | | mouse |
| ! ■ B1 | ILWE_AA_526 | 33.53 | 33.10 | 0.47 | 1.00 | | | eimeria |
| ! ■ B2 | ILWE_AA_526 | 32.61 | 33.10 | 0.47 | 1.00 | | | eimeria |
| ! ■ B3 | ILWE_AA_526 | 33.17 | 33.10 | 0.47 | 1.00 | | | eimeria |
| ! ■ B4 | ILWE_AA_526 | 25.49 | 25.32 | 0.23 | 1.00 | | | mouse |
| ! ■ B5 | ILWE_AA_526 | 25.06 | 25.32 | 0.23 | 1.00 | | | mouse |
| ! ■ B6 | ILWE_AA_526 | 25.41 | 25.32 | 0.23 | 1.00 | | | mouse |
| ! ■ B7 | ILWE_AA_534 | 32.20 | 31.96 | 0.25 | 1.00 | | | eimeria |
| ! ■ B8 | ILWE_AA_534 | 31.69 | 31.96 | 0.25 | 1.00 | | | eimeria |
| ! ■ B9 | ILWE_AA_534 | 31.99 | 31.96 | 0.25 | 1.00 | | | eimeria |
| ! ■ B10 | ILWE_AA_534 | 22.68 | 22.76 | 0.32 | 1.00 | | | mouse |
| ! ■B11 | ILWE_AA_534 | 23.11 | 22.76 | 0.32 | 1.00 | | | mouse |
| ! ■ B12 | ILWE_AA_534 | 22.49 | 22.76 | 0.32 | 1.00 | | | mouse |
| ! ■ C1 | ILWE_AA_637 | 14.82 | 15.75 | 2.02 | 1.00 | | | eimeria |
| ! | ILWE_AA_637 | 14.38 | 15.75 | 2.02 | 1.00 | | | eimeria |
| i | ILWE_AA_637 | 18.07 | 15.75 | 2.02 | 1.00 | | | eimeria |
| ! | ILWE_AA_637 | 23.66 | 23.13 | 0.56 | 1.00 | | | mouse |
| ! ■ C5 | ILWE_AA_637 | 23.18 | 23.13 | 0.56 | 1.00 | | | mouse |
| i∏ Ce | ILWE_AA_637 | 22.54 | 23.13 | 0.56 | 1.00 | | | mouse |
| ! □ C7 | ILWE_AA_535 | 30.78 | 31.00 | 0.21 | 1.00 | | | eimeria |
| i | ILWE_AA_535 | 31.20 | 31.00 | 0.21 | 1.00 | | | eimeria |
| i | ILWE_AA_535 | 31.02 | 31.00 | 0.21 | 1.00 | | | eimeria |
| ! ■ C10 | ILWE_AA_535 | 22.17 | 22.21 | 0.03 | 1.00 | | | mouse |
| ! ■ C11 | ILWE_AA_535 | 22.23 | 22.21 | 0.03 | 1.00 | | | mouse |



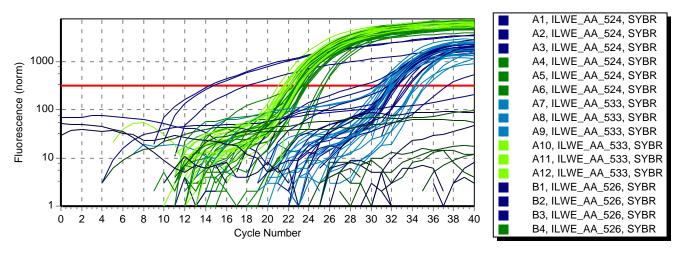
| Pos | Name | Ct SYBR | Ct Mean SYBR | Ct Dev. SYBR | Amount SYBR [Copies] | Amount Mean SYBR | Amount Dev. SYBR | Target SYBR |
|-----------------|-------------|---------|-----------------|-----------------|----------------------------|---------------------|---------------------|-------------|
| . C12 | ILWE_AA_535 | 22.23 | 22.21 | 0.03 | 1.00 | | | mouse |
| ! ■ D1 | ILWE_AA_660 | 31.68 | 30.39 | 1.34 | 1.00 | | | eimeria |
| ! ■ D2 | ILWE_AA_660 | 30.48 | 30.39 | 1.34 | 1.00 | | | eimeria |
| ! ■ D3 | ILWE_AA_660 | 29.00 | 30.39 | 1.34 | 1.00 | | | eimeria |
| ! | ILWE_AA_660 | 23.72 | 23.48 | 0.22 | 1.00 | | | mouse |
| ! ■ D5 | ILWE_AA_660 | 23.31 | 23.48 | 0.22 | 1.00 | | | mouse |
| i | ILWE_AA_660 | 23.40 | 23.48 | 0.22 | 1.00 | | | mouse |
| ! | ILWE_AA_536 | 35.19 | 34.94 | 0.64 | 1.00 | | | eimeria |
| ! ■ D8 | ILWE_AA_536 | 34.21 | 34.94 | 0.64 | 1.00 | | | eimeria |
| ! ■ D9 | ILWE_AA_536 | 35.42 | 34.94 | 0.64 | 1.00 | | | eimeria |
| ! ☐ D10 | ILWE_AA_536 | 23.09 | 23.09 | 0.05 | 1.00 | | | mouse |
| ! □ D11 | ILWE_AA_536 | 23.15 | 23.09 | 0.05 | 1.00 | | | mouse |
| ! ☐ D12 | ILWE_AA_536 | 23.05 | 23.09 | 0.05 | 1.00 | | | mouse |
| ! ■ E1 | ILWE_AA_525 | 32.47 | 32.27 | 0.24 | 1.00 | | | eimeria |
| ! ■ E2 | ILWE_AA_525 | 32.33 | 32.27 | 0.24 | 1.00 | | | eimeria |
| ! ■ E3 | ILWE_AA_525 | 32.01 | 32.27 | 0.24 | 1.00 | | | eimeria |
| ! ■ E4 | ILWE_AA_525 | 22.84 | 22.72 | 0.12 | 1.00 | | | mouse |
| ! ■ E5 | ILWE_AA_525 | 22.59 | 22.72 | 0.12 | 1.00 | | | mouse |
| ! ■ E6 | ILWE_AA_525 | 22.72 | 22.72 | 0.12 | 1.00 | | | mouse |
| ! □ ■ E7 | ILWE_AA_538 | 34.30 | 34.43 | 0.19 | 1.00 | | | eimeria |
| ! ■ E8 | ILWE_AA_538 | 34.35 | 34.43 | 0.19 | 1.00 | | | eimeria |
| ! ■ E9 | ILWE_AA_538 | 34.65 | 34.43 | 0.19 | 1.00 | | | eimeria |
| ! ■E10 | ILWE_AA_538 | 23.00 | 22.81 | 0.21 | 1.00 | | | mouse |
| ! □ E11 | ILWE_AA_538 | 22.87 | 22.81 | 0.21 | 1.00 | | | mouse |
| ! ■ E12 | ILWE_AA_538 | 22.58 | 22.81 | 0.21 | 1.00 | | | mouse |
| ! | ILWE_AA_641 | 31.92 | 31.93 | 0.16 | 1.00 | | | eimeria |
| ! ■ F2 | ILWE_AA_641 | 31.76 | 31.93 | 0.16 | 1.00 | | | eimeria |
| ! | ILWE_AA_641 | 32.09 | 31.93 | 0.16 | 1.00 | | | eimeria |
| ! | ILWE_AA_641 | 23.25 | 22.89 | 0.31 | 1.00 | | | mouse |
| ! | ILWE_AA_641 | 22.75 | 22.89 | 0.31 | 1.00 | | | mouse |
| ! | ILWE_AA_641 | 22.68 | 22.89 | 0.31 | 1.00 | | | mouse |
| ! | ILWE_AA_540 | 31.85 | 31.94 | 0.19 | 1.00 | | | eimeria |
| ! | ILWE_AA_540 | 31.81 | 31.94 | 0.19 | 1.00 | | | eimeria |
| ! | ILWE_AA_540 | 32.17 | 31.94 | 0.19 | 1.00 | | | eimeria |
| ! | ILWE_AA_540 | 21.70 | 21.70 | 0.03 | 1.00 | | | mouse |
| ! | ILWE_AA_540 | 21.74 | 21.70 | 0.03 | 1.00 | | | mouse |
| F12 | ILWE_AA_540 | 21.67 | 21.70 | 0.03 | 1.00 | | | mouse |
| • G1 | ILWE_AA_531 | 37.39 | 35.55 | 2.60 | 1.00 | | | eimeria |
| ! ∏ G 2 | ILWE_AA_531 | 33.72 | 35.55 | 2.60 | 1.00 | | | eimeria |



| Pos | Name | Ct SYBR | Ct Mean SYBR | Ct Dev. SYBR | Amount SYBR [Copies] | Amount Mean SYBR | Amount Dev. SYBR | Target SYBR |
|-----------------------|-------------|---------|-----------------|-----------------|----------------------------|---------------------|---------------------|-------------|
| <u>•</u> ■ G3 | ILWE_AA_531 | | 35.55 | 2.60 | 1.00 | | | eimeria |
| ! | ILWE_AA_531 | 22.23 | 22.15 | 0.14 | 1.00 | | | mouse |
| ! | ILWE_AA_531 | 21.99 | 22.15 | 0.14 | 1.00 | | | mouse |
| ! | ILWE_AA_531 | 22.23 | 22.15 | 0.14 | 1.00 | | | mouse |
| ! | ILWE_AA_541 | 32.03 | 31.71 | 0.60 | 1.00 | | | eimeria |
| ! ∏ G 8 | ILWE_AA_541 | 31.02 | 31.71 | 0.60 | 1.00 | | | eimeria |
| ! ∏ G 9 | ILWE_AA_541 | 32.08 | 31.71 | 0.60 | 1.00 | | | eimeria |
| _ !∏ | ILWE_AA_541 | 21.97 | 22.05 | 0.13 | 1.00 | | | mouse |
| _ ! | ILWE_AA_541 | 22.20 | 22.05 | 0.13 | 1.00 | | | mouse |
| ! | ILWE_AA_541 | 21.97 | 22.05 | 0.13 | 1.00 | | | mouse |
| - □ ■H1 | NTC | - | | | - | | | eimeria |
| - □ H 2 | NTC | - | | | - | | | eimeria |
| - □ ■H3 | NTC | - | | | - | | | eimeria |
| - □ ■H4 | NTC | - | | | - | | | mouse |
| - □ ■H5 | NTC | - | | | - | | | mouse |
| - □ ■H6 | NTC | - | | | - | | | mouse |
| - □ ■H7 | water | - | | | - | | | eimeria |
| - □ ■H8 | water | - | | | - | | | eimeria |
| - H 9 | water | - | | | - | | | eimeria |
| -TH10 | water | - | | | - | | | mouse |
| -T H11 | water | - | | | - | | | mouse |
| - □ ■H12 | water | - | | | - | | | mouse |



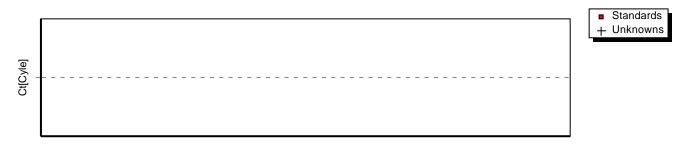
Amplification Plot



Threshold 310 (Noiseband)

Baseline automatic, Drift correction OFF

Standard curve



Amount[Copies]

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



Melting Curve SYBR

| Pos | Name | No. Tm SYBR | Tm x (°C) SYBR | Tm y (°C) SYBR | Mean SYBR | Dev. SYBR |
|----------------|-------------|-------------|----------------|----------------|-----------|-----------|
| <u>•</u> ■ A1 | ILWE_AA_524 | 0 | | | | _ |
| ! | ILWE_AA_524 | 0 | | | | |
| ! ■ A3 | ILWE_AA_524 | 0 | | | | |
| ! | ILWE_AA_524 | 1 | 79.9 | | | |
| ! | ILWE_AA_524 | 1 | 80.0 | | | |
| ! | ILWE_AA_524 | 1 | 79.9 | | | |
| ! | ILWE_AA_533 | 0 | | | | |
| ! ■ A8 | ILWE_AA_533 | 0 | | | | |
| ! | ILWE_AA_533 | 0 | | | | |
| ! | ILWE_AA_533 | 1 | 79.6 | | | |
| ! | ILWE_AA_533 | 1 | 79.7 | | | |
| ! | ILWE_AA_533 | 1 | 79.9 | | | |
| ! | ILWE_AA_526 | 0 | | | | |
| ! ■ B2 | ILWE_AA_526 | 0 | | | | |
| ! ■ B3 | ILWE_AA_526 | 0 | | | | |
| ! ■ B4 | ILWE_AA_526 | 1 | 79.9 | | | |
| ! | ILWE_AA_526 | 1 | 79.9 | | | |
| ! ■ B6 | ILWE_AA_526 | 1 | 79.9 | | | |
| ! | ILWE_AA_534 | 0 | | | | |
| ! ■ B8 | ILWE_AA_534 | 0 | | | | |
| ! ■ B9 | ILWE_AA_534 | 0 | | | | |
| ! ■ B10 | ILWE_AA_534 | 1 | 79.1 | | | |
| ! ■ B11 | ILWE_AA_534 | 1 | 79.2 | | | |
| ! ■ B12 | ILWE_AA_534 | 1 | 79.3 | | | |
| ! ☐ C1 | ILWE_AA_637 | 1 | 78.5 | | | |
| ! | ILWE_AA_637 | 1 | 78.3 | | | |
| i∏ C3 | ILWE_AA_637 | 0 | | | | |
| ! | ILWE_AA_637 | 1 | 79.8 | | | |
| ! | ILWE_AA_637 | 1 | 79.9 | | | |
| i∏ Ce | ILWE_AA_637 | 1 | 80.0 | | | |
| ! | ILWE_AA_535 | 0 | | | | |
| i | ILWE_AA_535 | 0 | | | | |
| i | ILWE_AA_535 | 0 | | | | |
| ! | ILWE_AA_535 | 1 | 78.7 | | | |
| ! ☐ C11 | ILWE_AA_535 | 1 | 78.8 | | | |
| ! ∏ C12 | ILWE_AA_535 | 1 | 79.0 | | | |
| | | | | | | |



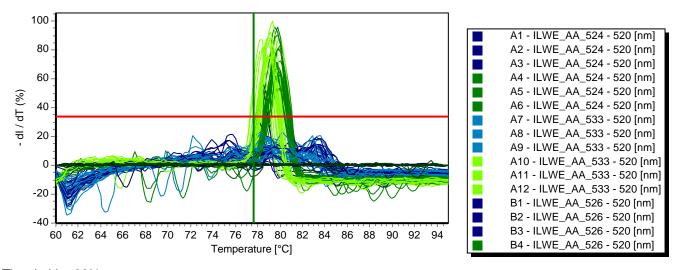
| Pos | Name | No. Tm SYBR | Tm x (°C) SYBR | Tm y (°C) SYBR | Mean SYBR | Dev. SYBR |
|-----------------|-------------|-------------|----------------|----------------|-----------|-----------|
| ! □ D1 | ILWE_AA_660 | 0 | | | | |
| ! □ D2 | ILWE_AA_660 | 0 | | | | |
| ! ∏ D3 | ILWE_AA_660 | 0 | | | | |
| ! ∏ D4 | ILWE_AA_660 | 1 | 79.5 | | | |
| ! □ D5 | ILWE_AA_660 | 1 | 79.6 | | | |
| . □ D6 | ILWE_AA_660 | 1 | 79.6 | | | |
| . □ D7 | ILWE_AA_536 | 0 | | | | |
| . □ D8 | ILWE_AA_536 | 0 | | | | |
| . □ D9 | ILWE_AA_536 | 0 | | | | |
| ! □ D10 | ILWE_AA_536 | 1 | 78.3 | | | |
| ! □ D11 | ILWE_AA_536 | 1 | 78.3 | | | |
| ! □ D12 | ILWE_AA_536 | 1 | 78.5 | | | |
| ! ■ E1 | ILWE_AA_525 | 0 | | | | |
| ! | ILWE_AA_525 | 0 | | | | |
| ! | ILWE_AA_525 | 0 | | | | |
| ! | ILWE_AA_525 | 1 | 79.5 | | | |
| ! ∏ E5 | ILWE_AA_525 | 1 | 79.8 | | | |
| i ∏ E6 | ILWE_AA_525 | 1 | 79.6 | | | |
| ! ∏ E7 | ILWE_AA_538 | 0 | | | | |
| ! | ILWE_AA_538 | 0 | | | | |
| ! | ILWE_AA_538 | 0 | | | | |
| ! ■ E10 | ILWE_AA_538 | 1 | 79.0 | | | |
| ! | ILWE_AA_538 | 1 | 79.0 | | | |
| ! ■ E12 | ILWE_AA_538 | 1 | 79.3 | | | |
| ! | ILWE_AA_641 | 0 | | | | |
| ! | ILWE_AA_641 | 0 | | | | |
| ! | ILWE_AA_641 | 0 | | | | |
| ! | ILWE_AA_641 | 1 | 79.6 | | | |
| ! | ILWE_AA_641 | 1 | 79.7 | | | |
| ! | ILWE_AA_641 | 1 | 79.8 | | | |
| ! ∏ F7 | ILWE_AA_540 | 0 | | | | |
| ! | ILWE_AA_540 | 0 | | | | |
| ! | ILWE_AA_540 | 0 | | | | |
| ! | ILWE_AA_540 | 1 | 78.8 | | | |
| ! ∏ F11 | ILWE_AA_540 | 1 | 78.9 | | | |
| ! | ILWE_AA_540 | 1 | 79.1 | | | |
| ! | ILWE_AA_531 | 0 | | | | |
| ! | ILWE_AA_531 | 0 | | | | |
| i G3 | ILWE_AA_531 | 0 | | | | |
| ! ∏ G4 | ILWE_AA_531 | 1 | 79.7 | | | |



| Pos | Name | No. Tm SYBR | Tm x (°C) SYBR | Tm y (°C) SYBR | Mean SYBR | Dev. SYBR |
|----------------|-------------|-------------|----------------|----------------|-----------|-----------|
| ! ∏ G5 | ILWE_AA_531 | 1 | 79.8 | | | |
| ! | ILWE_AA_531 | 1 | 79.9 | | | |
| ! | ILWE_AA_541 | 0 | | | | |
| ! | ILWE_AA_541 | 0 | | | | |
| ! | ILWE_AA_541 | 0 | | | | |
| ! | ILWE_AA_541 | 1 | 79.0 | | | |
| ! | ILWE_AA_541 | 1 | 79.0 | | | |
| ! | ILWE_AA_541 | 1 | 79.2 | | | |
| - ☐ H1 | NTC | 0 | | | | |
| - ∏ H2 | NTC | 0 | | | | |
| - ☐ H3 | NTC | 0 | | | | |
| - ☐ H4 | NTC | 0 | | | | |
| - ☐ H5 | NTC | 0 | | | | |
| - ∏ H6 | NTC | 0 | | | | |
| - ∏ H7 | water | 0 | | | | |
| - ∏ H8 | water | 0 | | | | |
| - □ H9 | water | 0 | | | | |
| - ∏ H10 | water | 0 | | | | |
| - ∏ H11 | water | 0 | | | | |
| - ☐ H12 | water | 0 | | | | |



Melting curve



Threshold 33%

