APPLICATION NOTE

Thermal cycler comparison: the ProFlex 96-Well PCR System demonstrates consistent, reliable performance

Introduction

The Applied Biosystems[™] ProFlex[™] PCR System was first introduced in 2013, and it has developed a strong reputation as a precise and reliable thermal cycler. This study compares the ProFlex 96-Well PCR System to our discontinued Applied Biosystems[™] GeneAmp[™] PCR System 9700 and other thermal cycler models, including:

- Bio-Rad[™] C1000 Touch[™] 96-Well Thermal Cycler
- MJ Research PTC-200 96-Well Thermal Cycler
- Eppendorf[™] Mastercycler[™] Pro S thermal cycler





Materials and methods

Two PCR templates (0.9 kb and 3 kb) were tested using the chemistries and amplification profiles shown in Figures 1 and 2.

| Product | Volume per reaction (µL) | Cat. No. |
|---------------------------------------------------------------------------------------|-----------------------------|----------|
| Invitrogen [™] Platinum [™] PCR SuperMix* | 46.0 | 11306016 |
| Forward primer (100 pmol/µL) | 0.5 | A15612 |
| Reverse primer (100 pmol/µL) | 0.5 | A15612 |
| DNA template (50 ng plasmid) | 1.0 | NA |
| Invitrogen [™] UltraPure [™] DNase/RNase-Free Distilled Water | 2.0 | 10977015 |

Figure 1. PCR reaction composition and cycling conditions for the 0.9 kb template.



Figure 2. PCR reaction composition and cycling conditions for the 3 kb template.

Invitrogen[™] E-Gel[™] Agarose Gels with SYBR[™] Safe DNA Gel Stain, 1.2% (Cat. No. G521801) and TrackIt[™] 1 Kb Plus DNA Ladder (Cat. No. 10488085) were used to visualize PCR products. To account for variation in the thermal block, PCR reactions were placed in the positions shown in Figure 3, with a negative control placed at position G12.



Figure 3. PCR reaction positions in the thermal block.

Results

PCR amplification was comparable across the five instruments tested, for the 0.9 kb template (Figure 4). There was no major difference between the instruments in terms of product yield. Each instrument generated the correctly sized PCR product in all wells tested. However, for the 3 kb template (Figure 5), the Bio-Rad C1000 Touch 96-Well Thermal Cycler showed positional variability.



Figure 4. Amplification of the 0.9 kb template.

Figure 5. Amplification of the 3 kb template.

Conclusions

The ProFlex PCR System demonstrated product yield comparable to that of the proven and reliable GeneAmp PCR System 9700. Significantly less well-to-well variability was observed with these systems, compared to the Bio-Rad C1000 Touch 96-Well Thermal Cycler, with the 3 kb template. These results are assay-specific, and results may vary depending on the PCR experimental conditions.

Instrument specifications

| | ProFlex PCR System | GeneAmp PCR System 9700 | Bio-Rad C1000 Touch 96-Well Thermal Cycler | Eppendorf Mastercycler Pro S |
|-----------------------------------|----------------------------------------------------------------------------------------------------------------------------------------------------|---------------------------------------------------------------------------------------------------------------------------|-----------------------------------------------------------------------------------|---------------------------------------|
| Formats available | 3 x 32-well independent control; 1 x 96-well VeriFlex [™] Block; 2 x 96- well; dual flat block for chip formats; 2 x 384-well | 1 x 96-well, 0.2 mL; 1 x 60-well, 0.5 mL; 2 x 96-well, 0.2 mL; dual flat block for chip formats; 2 x 384-well | 1 x 96-well, 0.2 mL; 1 x 96 deep well; 2 x 48-well, 0.2 mL; 1 x 384-well | 1 x 96-well, 0.2 mL; 1 x 384-well |
| Maximum block ramp rate | 6°C/sec | 2.6°C/sec (for gold-plated block) | 5°C/sec | Approx. 6°C/sec |
| Maximum sample ramp rate | 4.4°C/sec (measured at 1 µL volume) | 2.3°C/sec (for gold-plated block; measured at 1 µL volume) | Not published | Not published |
| Temperature range | 0°–100°C | 4°-99°C | 0°–100°C | 4°-99°C |
| Temperature accuracy | ±0.25°C (35°–99.9°C) | ±0.25°C (35°–99.9°C) | ±0.2°C of programmed target at 90°C | ±0.2°C |
| Temperature uniformity | <0.5°C (20 sec after reaching 95°C) | ±0.5°C (30 sec after reaching 95°C) | ± 0.4 °C well-to-well within 10 sec of arrival at 90 °C | ±0.3°C (20°–72°C) ±0.4°C (95°C) |
| Input power | 100–240 V, 50–60 Hz Max: 950 VA | 100–240 V, 50–60 Hz Max: 725 VA | Max: 850 W | Max: 950 W |
| Display | 8.4 in. color TFT LCD touch screen | Monochrome LCD display and tactile keypad | 8.5 in. LCD display and touch screen | Removable, push-button control panel |
| Ports | USB x 3 | PCMCIA | USB A x 5, USB B x 1 | USB, CAN |
| Memory | USB, onboard with >1,000 protocols | PCMCIA, onboard | USB, onboard with >1,000 protocols | Not published |
| Approx. dimensions (W x D x H) | 33 x 57 x 27 cm (13 x 22 x 11 in.) | 30 x 41 x 26 cm (12 x 16 x 10 in.) | 33 x 46 x 20 cm (13 x 18 x 8 in.) | 26 x 42 x 37 cm (10 x 16 x 15 in.) |
| Approx. weight | 19 kg (41 lb) | 9 kg (19 lb) | 10 kg (23 lb) | 19 kg (41 lb) |
| Temperature optimization range | VeriFlex Blocks: 5°C in 32-well blocks (2 zones); 25°C in 96-well blocks (6 VeriFlex zones) | NA | 30°–100°C gradient block | 30°–99°C gradient block |
| Programming options | Touch screen, and preloaded onboard protocols | Buttons | Step-based graphical and automatic | Buttons |
| Security features | 3 levels of security for regulated environments | NA | Optional log-in required; mode for regulated environments | Safe mode available |
| VeriFlex Blocks | Yes | No | No | No |
| External software | Mobile application for remote monitoring | External PC-based software available | External PC-based software available | External PC-based software available |

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Ordering information

| Product | Quantity | Cat. No. |
|-------------------------------------------------------|------------------|----------|
| ProFlex 96-Well PCR System | 1 system | 4484075 |
| ProFlex 3 x 32-Well PCR System | 1 system | 4484073 |
| ProFlex 2 x 96-Well PCR System | 1 system | 4484076 |
| ProFlex 2 x Flat PCR System | 1 system | 4484078 |
| ProFlex 2 x 384-Well System | 1 system | 4484077 |
| ProFlex 96-Well Sample Block | 1 block | 4483637 |
| ProFlex 3 x 32-Well Sample Block | 1 block | 4483638 |
| ProFlex 2 x 96-Well Sample Block | 1 block | 4484071 |
| ProFlex 2 x Flat Sample Block | 1 block | 4484074 |
| ProFlex 2 x 384-Well Sample Block | 1 block | 4484072 |
| Platinum PCR SuperMix High Fidelity | 100 reactions | 12532016 |
| DNA oligo (custom), 25 nmol, desalted, dry | 25 nmol | A15612 |
| UltraPure DNase/RNase-Free Distilled Water | 500 mL | 10977015 |
| E-Gel Agarose Gels with SYBR Safe DNA Gel Stain, 1.2% | 18 gels | G521801 |
| TrackIt 1 Kb Plus DNA Ladder | 100 applications | 10488085 |
| MicroAmp Optical 96-Well Reaction Plate with Barcode | 20 plates | 4306737 |
| MicroAmp Clear Adhesive Film | 100 films | 4306311 |

Find out more at **thermofisher.com/proflex**



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