



PCR

VeritiPro Dx Thermal Cycler—technical comparison to the Veriti Dx Thermal Cycler

Introduction

The Applied Biosystems™ Veriti™ Dx Thermal Cycler set a new standard for innovation when it was launched in 2011. It was the first and only thermal cycler for *in vitro* diagnostic use, classified as a US FDA class I medical device and conforming to IVDD requirements in Europe. In June 2023, the Veriti Dx Thermal Cycler was upgraded to the Applied Biosystems™ VeritiPro™ Dx Thermal Cycler, which is CE-IVD labeled/IVDR-compliant and classified as a US FDA class I medical device. The VeritiPro Dx Thermal Cycler has an innovative design and features the latest

block technology. These advancements can help provide high ramp rates, quiet runs, and a small footprint while continuing to offer the same premium performance (Figure 1). This study demonstrates that the VeritiPro Dx Thermal Cycler delivers equivalent reliability, accuracy, and consistency in amplification uniformity as the Veriti Dx Thermal Cycler, by comparing coefficients of variation (CV) and robustness across a variety of PCR consumables.

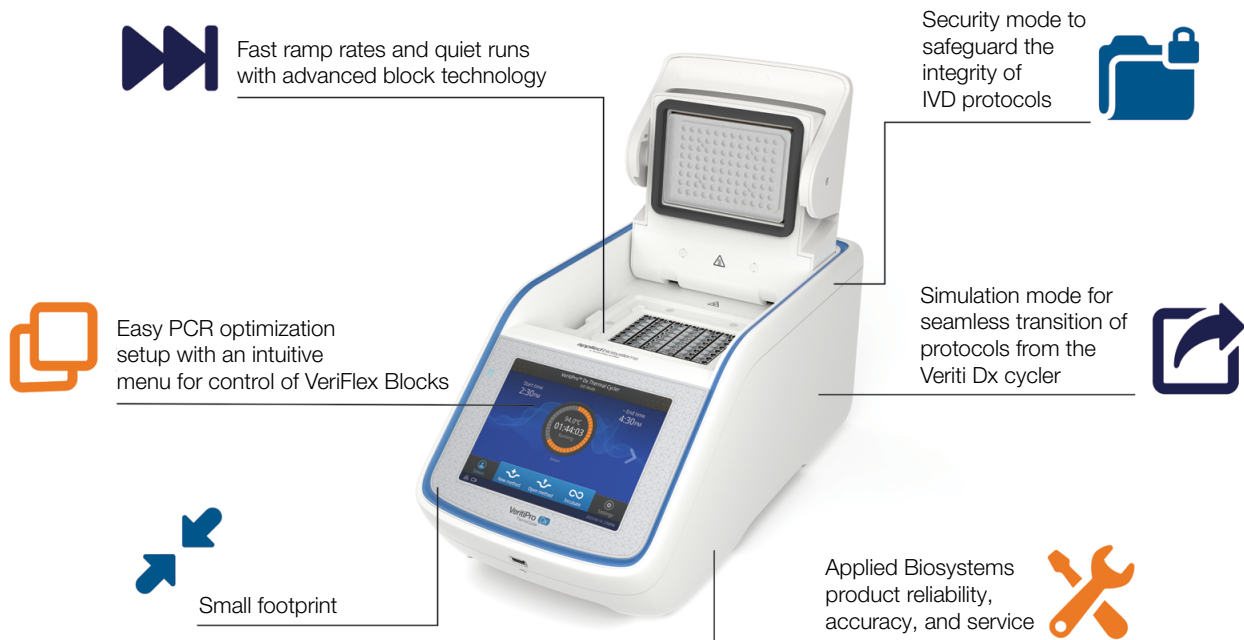


Figure 1. The innovative design and advanced features of the VeritiPro Dx Thermal Cycler.

Highlights of the VeritiPro Dx Thermal Cycler (CE-IVD, IVDR-compliant)

The VeritiPro Dx Thermal Cycler is an *in vitro* diagnostic (IVD) end-point thermal cycler designed to amplify nucleic acids from human-derived specimens using polymerase chain reaction (PCR). The VeritiPro Dx Thermal Cycler provides two security modes that help to safeguard the integrity of validated protocols and experimental results. The Applied Biosystems™ VeriFlex™ Blocks offer precise control over 6 independent temperature zones for accurate PCR optimization. The expanded 8-inch touchscreen and improved user interface provide an intuitive, interactive system for easy programming. Protocol setup includes an option for simulation mode, supports convenient transition of protocols, mimics the ramp rate of the Veriti Dx Thermal Cycler, and eliminates the need for re-optimization. The instrument was also designed with improved vent technology to reduce noise for an enhanced customer experience. These updates are included in a more compact instrument that can easily fit in a small workspace (Table 1).

Materials and methods

To verify the performance of the VeritiPro Dx Thermal Cycler, a variety of Applied Biosystems™ PCR plastics were used during testing, which was conducted at an ambient temperature within 15–30°C with relative humidity of 15–80% under noncondensing conditions. A complete list of Applied Biosystems™ MicroAmp™

consumables used in this study is shown in Table 2. All PCR plate consumables were tested using the Applied Biosystems™ Veriti™ Dx 96-Well Thermal Cycler, 0.2 mL (Cat. No. 4452300) and the VeritiPro Dx Thermal Cycler (Cat. No. A55826). For amplification uniformity testing, a single bulk reaction was prepared using Applied Biosystems™ Power SYBR™ Green PCR Master Mix (Cat. No. 4367660) according to the standard protocol. The Lambda DNA standard (Component C from the Invitrogen™ Quant-iT™ PicoGreen™ dsDNA Assay Kit, Cat. No. P7589) was used as a template at a final concentration of 0.01 ng/μL with lambda forward primer (5'-GATGAGTTCGTGTCGGTACAAC-3') and lambda reverse primer (5'-ACGGCTGCACGGAGTTCAGTATG-3') at a concentration of 0.2 μM each. The bulk reaction was tested at a 10 μL or 100 μL volume for 96-well reactions using different PCR consumables on the Veriti Dx and VeritiPro Dx Thermal Cyclers. The following thermal profile was used: 95.0°C for 10 min; 25 cycles at 94.0°C for 15 sec and 69.5°C for 90 sec; and a final stage at 72.0°C for 7 min. Upon completion of thermal cycling, the PCR reactions were transferred to an Applied Biosystems™ QuantStudio™ 5 Real-Time PCR System, 96-well, 0.2 mL (Cat. No. A28139) for fluorometric analysis. Amplification uniformity was calculated as a CV using the following formula:

$$CV = \frac{\text{Standard deviation of SYBR Green signal for each run}}{\text{Average SYBR Green signal for each run}} \times 100\%$$

Table 1. Specifications comparison between the Veriti Dx and VeritiPro Dx Thermal Cyclers.

	Veriti Dx Thermal Cycler	VeritiPro Dx Thermal Cycler
Block format	96-well, 0.2 mL aluminum block	96-well, 0.2 mL alloy block
Maximum block ramp rate	3.9°C/sec	6.0°C/sec
Temperature accuracy	±0.25°C (35–99°C)	±0.25°C (35–99.9°C)
Temperature range	0–100.0°C	0–100°C
Temperature uniformity	<0.5°C (20 sec after reaching 95°C)	<0.5°C (30 sec after reaching 95°C)
Reaction volume range	10–100 μL	10–100 μL
VeriFlex Blocks range	25°C range across blocks, 6 temperature zones (up to 5°C per zone)	25°C range across blocks, 6 temperature zones (up to 5°C per zone)
Display	6.5 in. VGA 32K color touchscreen	8 in. color TFT LCD touchscreen
Maximum fan noise	Operation: <61.3 dBA; idling: <57.1 dBA	Operation: <53 dBA; idling: <40 dBA
Dimensions (H x W x D)	24.5 x 23.7 x 48.5 cm	21.7 x 24.5 x 46.5 cm

Table 2. PCR consumables used in amplification uniformity testing.

Product	Cat. No.
MicroAmp Optical 96-Well GPLE Reaction Plates with Barcode	4481192
MicroAmp EnduraPlate Optical 96-Well Clear GPLE Reaction Plates with Barcode	4483348
MicroAmp Clear Adhesive Film	4306311
MicroAmp Optical Adhesive Covers GPLE	A49767
MicroAmp 8-Tube Strip, 0.2 mL	N8010580
MicroAmp 8-Cap Strip, clear	N8010535
MicroAmp Optical 8-Cap Strip	4323032
MicroAmp Reaction Tube with Cap, 0.2 mL	N8010540
MicroAmp Reaction Tube without Cap, 0.2 mL	N8010533
MicroAmp Optical 8-Tube Strip with Attached Optical Caps, 0.2 mL	A30588
MicroAmp 8-Tube Strip with Attached Domed Caps, 0.2 mL	A30589

For robustness testing, short-amplicon reactions were prepared using Applied Biosystems™ AmpliTaq Gold™ 360 Master Mix (Cat. No. 4398886) or Applied Biosystems™ AmpliTaq Gold™ Fast PCR Master Mix (Cat. No. 4390941) and human genomic DNA (Roche, Cat. No. 11691112001), according to the standard protocols.

Long-amplicon reactions were prepared using the Applied Biosystems™ SequelPrep™ Long PCR Kit with dNTPs (Cat. No. A10498) according to the standard protocol. Four-plex 10 µL PCR reactions were prepared using Invitrogen™ Platinum™ SuperFi™ II Green PCR Master Mix, to compare different PCR consumables on the Veriti Dx and VeritiPro Dx Thermal Cyclers. Following PCR, gel electrophoresis was performed with the Invitrogen™ E-Gel™ Power Snap Electrophoresis Device (Cat. No. G8100) to separate amplicons according to size. Gels were visualized using the Invitrogen™ iBright™ FL1500 Imaging System (Cat. No. A44241).

Results

Amplification uniformity

CVs were calculated using fluorescence intensity data from one Veriti Dx Thermal Cycler and three VeritiPro Dx Thermal Cyclers. Figure 2 shows amplification uniformity across a 96-well plate for two reaction volumes, 10 µL (low) and 100 µL (high). Figure 3 shows thermal cycler uniformity evaluated across various combinations of PCR plates, adhesive film, tube strips, and cap strips by measuring the average fluorescence intensity of 10 µL (for 96-well) PCR reactions. Run-to-run amplification uniformity on a 96-well plate is demonstrated in Figure 4, showing the VeritiPro Dx Thermal Cycler provides reproducible thermal performance across separate 10 µL PCR reactions. Overall, results show comparable well-to-well and run-to-run uniformity between thermal cyclers using different reaction volumes and PCR consumables. Results were reproducible across the three VeritiPro Dx Thermal Cyclers.

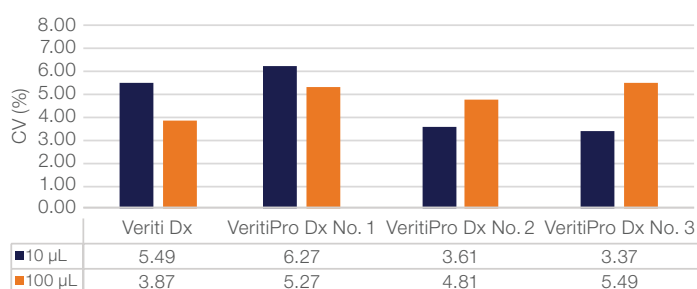


Figure 2. Comparison of well-to-well amplification uniformity between one Veriti Dx and three VeritiPro Dx Thermal Cyclers at low (10 µL) and high (100 µL) reaction volumes. The CV for each run was calculated using fluorescence intensity data from PCR using an Applied Biosystems™ MicroAmp™ Optical 96-Well Reaction Plate sealed with Applied Biosystems™ MicroAmp™ Clear Adhesive Film.

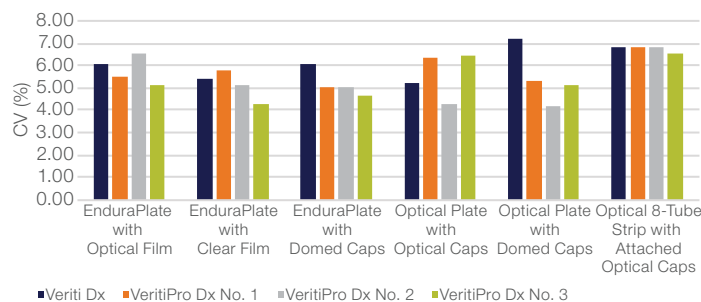


Figure 3. Comparison of well-to-well amplification uniformity between one Veriti Dx 96-Well Thermal Cycler and three VeritiPro Dx Thermal Cyclers using various combinations of PCR consumables, including MicroAmp EnduraPlate and Optical plates. The CV for each run was calculated using fluorescence intensity data from low-volume (10 µL) reactions.

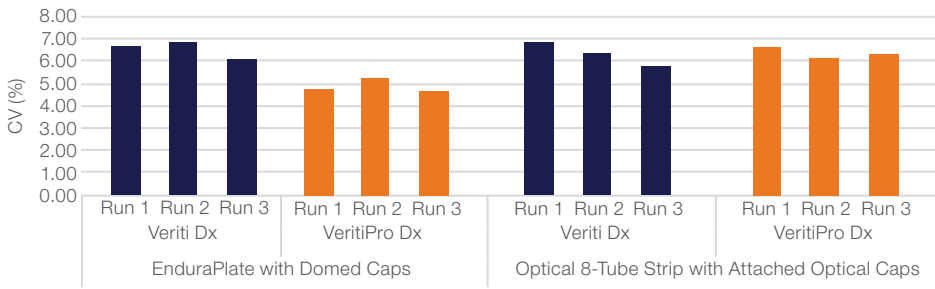


Figure 4. Comparison of run-to-run amplification uniformity between Veriti Dx and VeritiPro Dx Thermal Cyclers. The CV for each run was calculated using fluorescence intensity data from 10 μ L reactions.

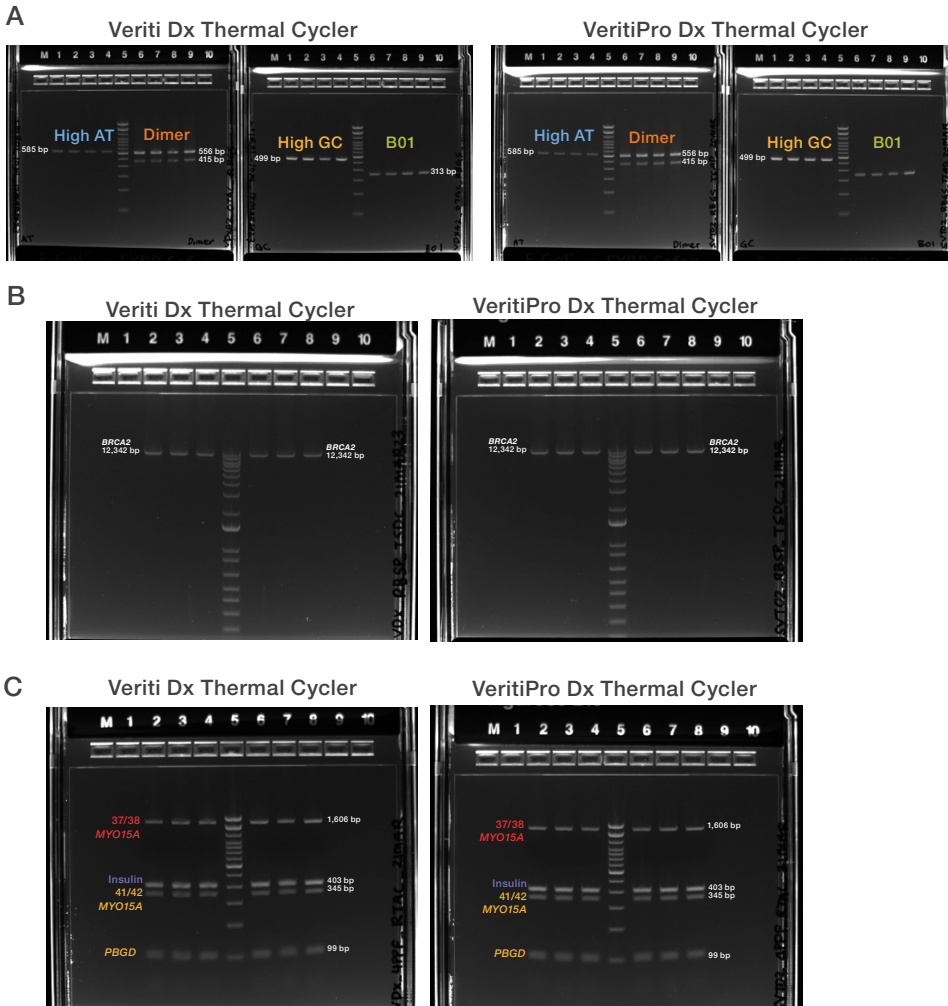


Figure 5. Comparison of robustness between Veriti Dx and VeritiPro Dx Thermal Cyclers. (A) AmpliAq Gold 360 Master Mix was used to amplify single, short human genomic DNA fragments (313 bp, 499 bp, 556 bp, or 585 bp) of different complexities in Applied Biosystems™ MicroAmp™ 8-Tube Strips with Attached Domed Caps. B01 denotes the breast cancer type 1 susceptibility protein isoforms 2 and 3. (B) The SequelPrep Long PCR Kit was used to amplify a single, long human genomic DNA amplicon (12,342 bp, *BRCA2* gene) in MicroAmp 8-Tube Strips with Applied Biosystems™ MicroAmp™ 8-Cap Strips. (C) Platinum SuperFi II Green PCR Master Mix was used to simultaneously amplify four DNA amplicons (99 bp, 345 bp, 403 bp, and 1,606 bp) in MicroAmp Reaction Tubes with Caps. Both 37/38 and 41/42 denote the unconventional myosin XV gene *MYO15A*. *PBGD* denotes porphobilinogen deaminase isoforms 1 and 2.

Robust thermal performance

To demonstrate robustness, the Veriti Dx and VeritiPro Dx Thermal Cyclers were tested by amplifying targets of different lengths and complexities, including standard DNA sequences, high AT- or GC-rich sequences, and amplicons likely to form dimers. Four-plex PCR was performed to verify the capability of the VeritiPro Dx Thermal Cycler to amplify targets of different lengths simultaneously in a single reaction. Successful amplification of the targets was demonstrated by the presence of distinct bands of correct sizes after gel electrophoresis. Results in Figure 5 indicate that the VeritiPro Dx Thermal Cycler provides thermal robustness equivalent to that of the Veriti Dx Thermal Cycler without the need to re-optimize experiments.

Easy protocol transition

For users concerned about re-optimizing protocols to run on the VeritiPro Dx Thermal Cycler, the simulation mode eliminates the need to do this work. This mode allow the user to run protocols under the same conditions as a different thermal cycler. Users can choose the VeritiPro Dx Thermal Cycler for easy instrument transition and consistency in experimental protocols.

Security mode for IVD protocols

For users concerned about safeguarding the integrity of validated protocols and results, there are two security modes under the administrative profile.

- Creation mode: enables users to view, modify, and run all methods
- Run-only mode: restricts user access to the IVD folder and permits users to only view and run IVD methods; this helps to reduce the risk of unauthorized use and accidental or intentional misuse

Discussion

Achieving consistent amplification uniformity and robust thermal performance during PCR are crucial factors to obtaining high-quality results. In this study, we have shown that the VeritiPro Dx Thermal Cycler provides thermal capabilities equivalent to those of the Veriti Dx Thermal Cycler for uniformity and robustness, with templates of differing lengths and complexities.

Results from the VeritiPro Dx Thermal Cycler were consistent across multiple combinations of PCR plates, tubes, strip tubes, adhesive films, and cap strips. Both low (10 μ L) and high (100 μ L) reaction volumes were tested on the VeritiPro Dx Thermal Cycler using the same experimental protocols that have been optimized and verified on the Veriti Dx Thermal Cycler, without the need to re-optimize. Additionally, the VeritiPro Dx Thermal Cycler offers higher ramp rates, allowing for faster time-to-results.

Ordering information

Product	Cat. No.
VeritiPro Dx Thermal Cycler with IQ/OQ	A57751
VeritiPro Dx Thermal Cycler with IQ/OQ, Extended 1-Year Warranty, and Annual PM with OQ	A57752
MicroAmp EnduraPlate Optical 96-Well Clear GPLE Reaction Plates with Barcode	4483348
MicroAmp Optical 96-Well GPLE Reaction Plates with Barcode	4481192
MicroAmp Clear Adhesive Film	4306311
Optical Adhesive Covers GPLE	A49767
MicroAmp 96-Well Tray for VeriFlex Blocks	4379983
MicroAmp 96-Well Tray/Retainer Set	4381850
MicroAmp Optical 8-Tube Strip with Attached Optical Caps, 0.2 mL	A30588
MicroAmp 8-Tube Strip with Attached Domed Caps, 0.2 mL	A30589
MicroAmp 8-Cap Strip, clear	N8010535
MicroAmp 8-Tube Strip, 0.2 mL	N8010580
MicroAmp Reaction Tube with Cap, 0.2 mL	N8010540
MicroAmp Reaction Tube without Cap, 0.2 mL	N8010533

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For *In Vitro* Diagnostic Use. The VeritiPro Dx Thermal Cycler is available in the US, Europe, and other selected countries globally. Please check with your Thermo Fisher Scientific representative for availability in your country.

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