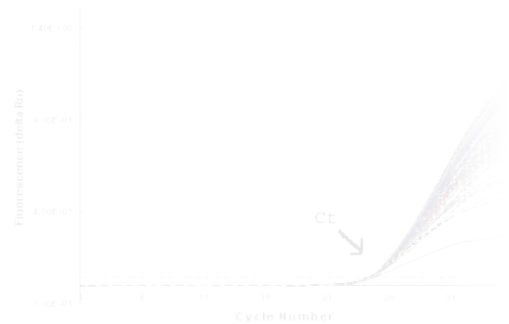


high-performance  
**hard-frame PCR plates**

RigidAuto PCR Plates



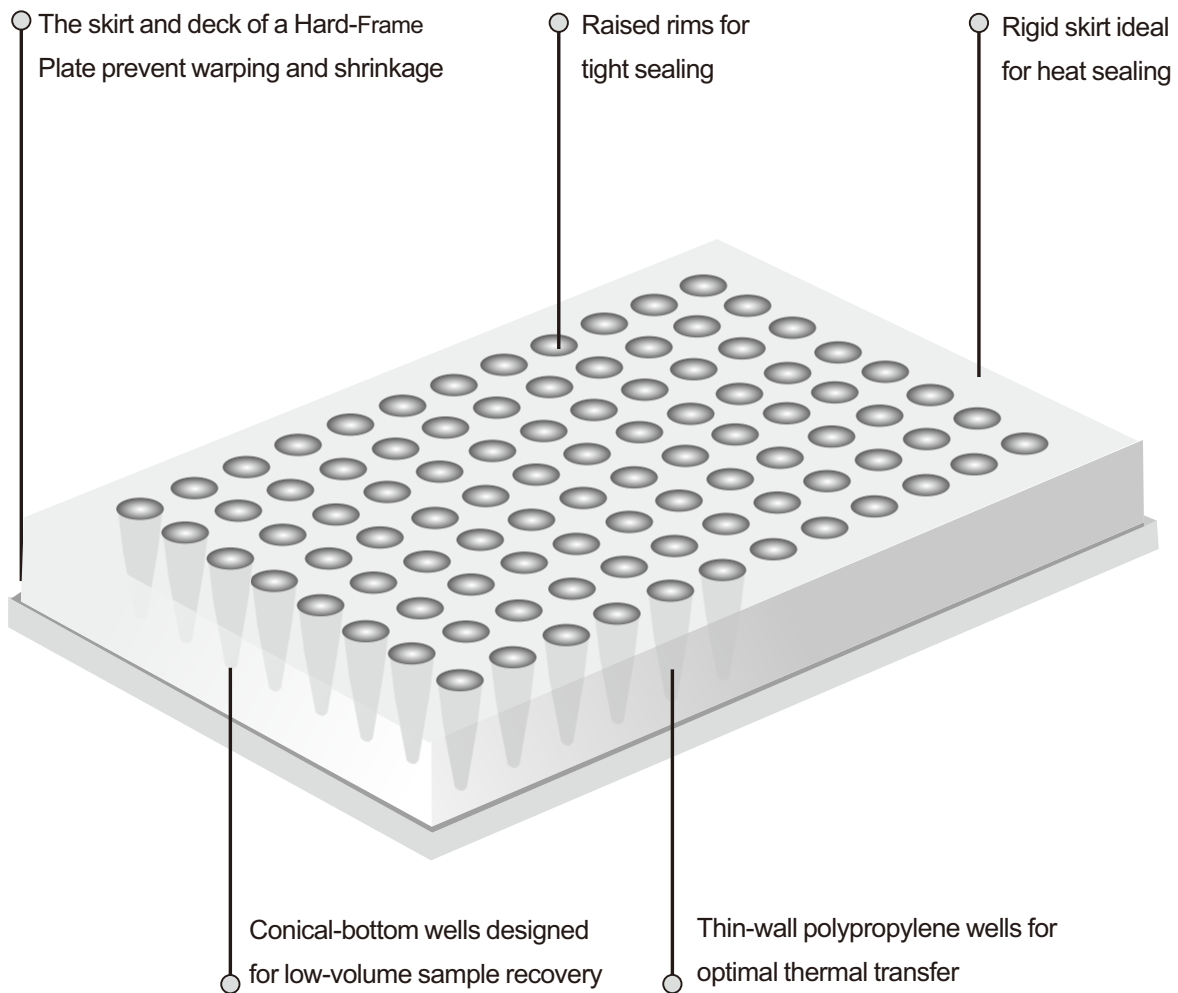


## The ultimate choice for high-throughput PCR and automated handling

Hard-Frame PCR Plates are engineered to endure the rigorous demands of thermal cycling, robotic automation, and heat sealing, representing an advanced innovation in PCR plate design. The skirt and deck are manufactured from a durable, thermostable polymer, eliminating warping issues during robotic handling. This robust frame ensures dimensional stability, preventing the distortion and shrinkage commonly observed in standard single-component polypropylene PCR plates under high-temperature thermal cycling or heat sealing. Ultra-thin well walls and inter-well uniformity ensure optimal compatibility and stable heat transfer. Each well features raised rims to facilitate secure sealing across multiple methods, including pressure-sensitive films, adhesive seals, and heat-sealing technologies.

### Product Features

<b>Material</b>	Polycarbonate (frame), polypropylene (wells)
<b>Resistance to chemicals</b>	The plates including border show a high resistance to UV light and chemicals
<b>Dimensions</b>	ACC. to ANSI/SLAS 1-2004, ANSI/SLAS 3-2004 and ANSI/SLAS 4-20041
<b>Operational temperature</b>	-80 °C to +120 °C
<b>Autoclavability</b>	Autoclavable (121°C, 20 min), open status. The stability of the single-use items can be compromised
<b>Type</b>	96 wells, 384 wells
<b>Max. centrifugation stability</b>	2,250 xg
<b>low adsorption</b>	Customizable
<b>barcoded</b>	Customizable
<b>Sterilization</b>	By default, it does not sterilize. Sterilization can be customized. For specific details, please consult the salesperson
<b>Colour</b>	Customizable



**Hard-frame PCR plates are specifically designed for high-performance thermal cycling applications, offering optimal thermal conductivity and structural integrity in:**

◆ **High-Throughput Applications:**

Next-generation sequencing (NGS) library preparation  
 Large-scale genotyping studies  
 Population-scale SNP screening

◆ **Advanced Research Applications:**

Digital PCR workflows  
 Multiplex PCR reactions  
 Low-volume ( $\leq 10\mu\text{L}$ ) amplification protocols

◆ **Precision Molecular Diagnostics:**

Clinical qPCR testing  
 Pathogen detection assays  
 Liquid biopsy workflows

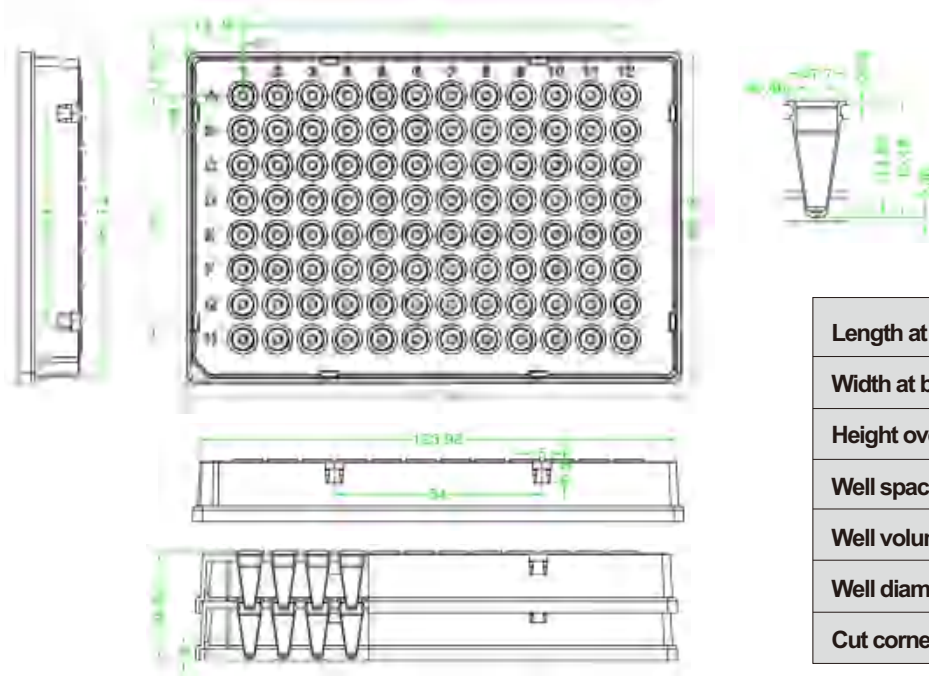
◆ **Automated Laboratory Systems:**

Robotic liquid handling platforms  
 Automated sample-to-answer systems  
 High-density array processing

## 200 $\mu$ L 96-well full skirted PCR plate (type B)



- ◆ Full-skirted design. Cut corner: H1
- ◆ Compatible with Bio-Rad instruments and most high-throughput automation platforms
- ◆ Available with low adsorption and customizable barcoding



Length at base plane	127.76 mm
Width at base plane	85.48 mm
Height overall	16.1 mm
Well spacing	9 mm
Well volume	200 $\mu$ L
Well diameter at opening	5.46 mm
Cut corner	H1

### Ordering information

Part No.	Description	Package
PCRP-20BFS-C2	200 $\mu$ L 96-well Full skirt, black print, clear base, clear well	10 pcs/box, 10 boxes/case
PCRP-20BFS-W1	200 $\mu$ L 96-well Full skirt, black print, white base, clear well	10 pcs/box, 10 boxes/case
PCRP-20BFS-W2	200 $\mu$ L 96-well Full skirt, black print, white base, white well	10 pcs/box, 10 boxes/case

### Recommended Instrument Compatibility (Continuously Updated)

#### Bio-Rad:

CFX96™, 1000, DNA Engine Opticon®, Chromo4™, PTC-100®, DNA Engine®

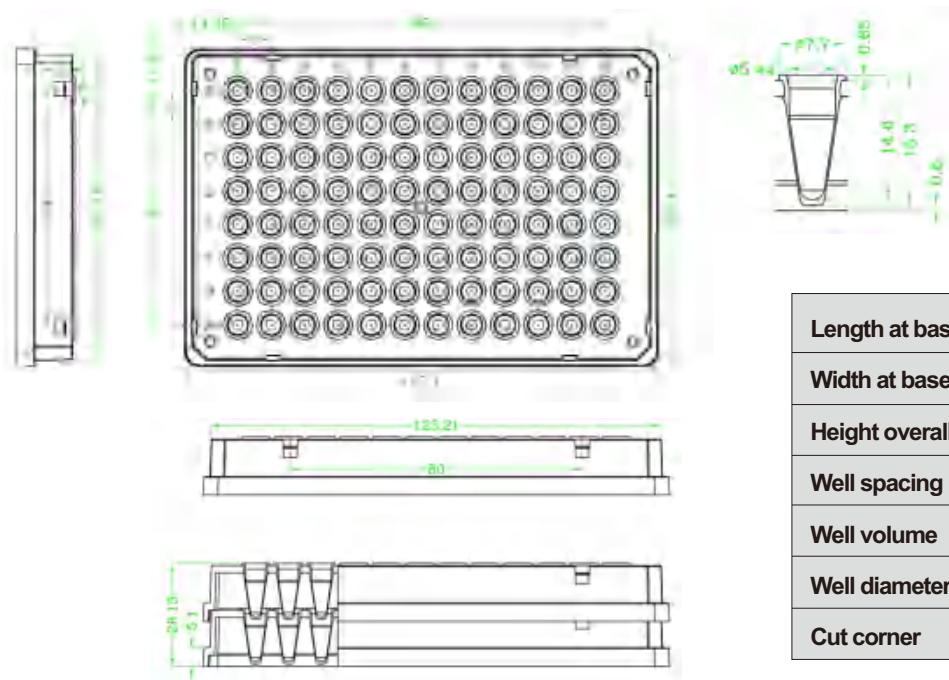
#### Eppendorf:

MasterCycler Series

## 200 $\mu$ L 96-well full skirted PCR plate (Type E)



- ◆ Full skirted design. Cut corner: H1/A12/H12
- ◆ Compatible with most PCR/RT-PCR thermal cyclers and Eppendorf instruments
- ◆ Available with low adsorption and customizable barcoding



Length at base plane	127.7 mm
Width at base plane	85.4 mm
Height overall	16.1 mm
Well spacing	9 mm
Well volume	200 $\mu$ L
Well diameter at opening	5.44 mm
Cut corner	H1/A12/H12

### Ordering information

Part No.	Description	Package
PCRP-20EFS-C2	200 $\mu$ L 96-well Full skirt, black print, clear base, clear well	10 pcs/box, 10 boxes/case
PCRP-20EFS-W1	200 $\mu$ L 96-well Full skirt, black print, white base, clear well	10 pcs/box, 10 boxes/case

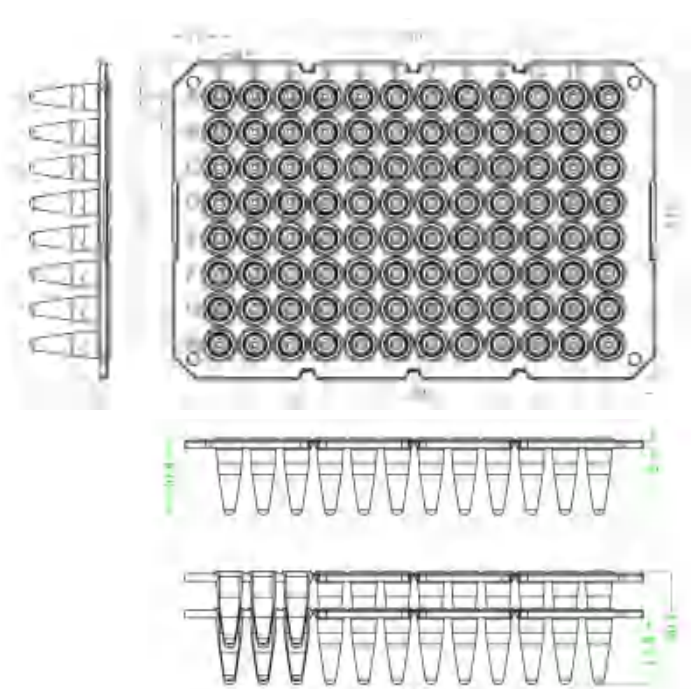
### Recommended Instrument Compatibility (Continuously Updated)

**Eppendorf:**  
MasterCycler Series

## 200 $\mu$ L 96-well unskirted PCR plate (type E)



- ◆ Unskirted design. Cut corner: A1/H1/A12/H12
- ◆ Conveniently precut into 24- or 48-well segments
- ◆ Compatible with most PCR/RT-PCR thermal cyclers and Eppendorf instruments
- ◆ Available with low adsorption



Length at base plane	123 mm
Width at base plane	81 mm
Height overall	20.4 mm
Well spacing	9 mm
Well volume	200 $\mu$ L
Well diameter at opening	5.44 mm
Cut corner	A1/H1/A12/H12

### Ordering information

Part No.	Description	Package
PCRP-20ETNS-C2	200 $\mu$ L 96-well unskirted, black print, clear base, clear well (Conveniently precut into 24- or 48-well segments)	10 pcs/box, 10 boxes/case

### Recommended Instrument Compatibility (Continuously Updated)

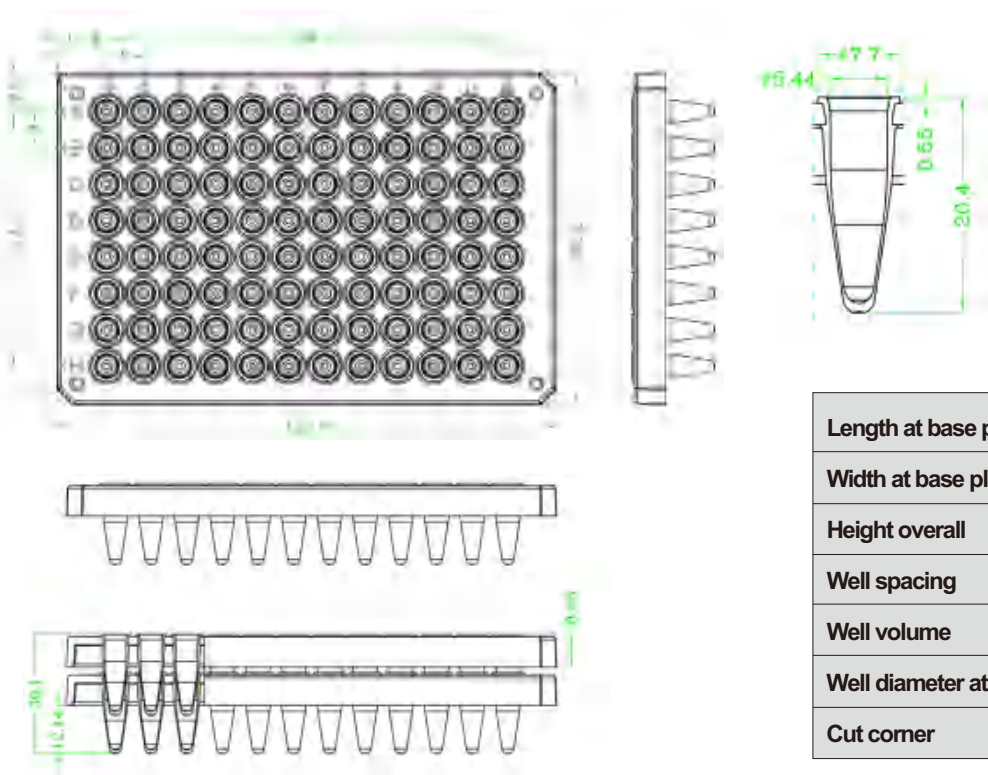
#### Eppendorf:

MasterCycler Series

## 200 $\mu$ L 96-well half skirt PCR plate (type E)



- ◆ Half skirted design. Cut corner: H1/A12/H12
- ◆ Compatible with most PCR/RT-PCR thermal cyclers and Eppendorf instruments
- ◆ Available with low adsorption and customizable barcoding



Length at base plane	123.6 mm
Width at base plane	82.2 mm
Height overall	20.4 mm
Well spacing	9 mm
Well volume	200 $\mu$ L
Well diameter at opening	5.44 mm
Cut corner	A1/A12/H12

### Ordering information

Part No.	Description	Package
PCRP-20EHS-C2	200 $\mu$ L 96-well Half skirted, black print, clear base, clear well	10 pcs/box, 10 boxes/case
PCRP-20EHS-C1	200 $\mu$ L 96-well Half skirted, black print, clear base, white well	10 pcs/box, 10 boxes/case

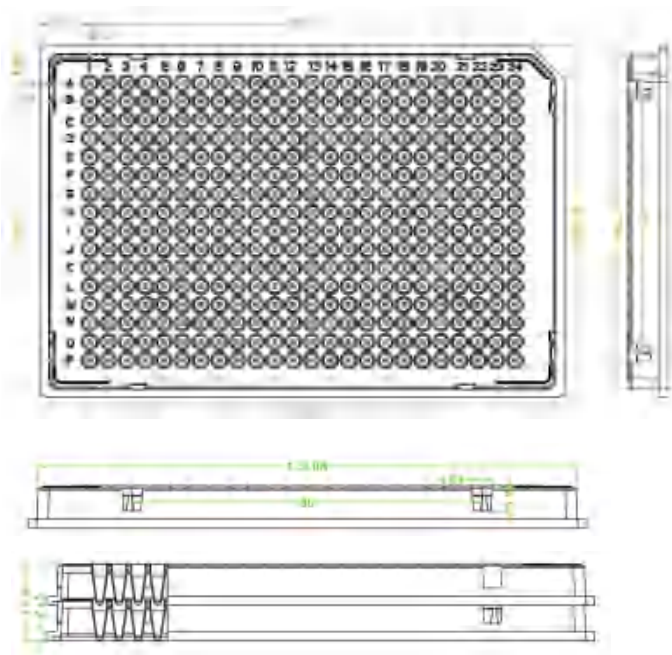
### Recommended Instrument Compatibility (Continuously Updated)

**Eppendorf:**  
MasterCycler Series

## 40 $\mu$ L 384-well PCR plate (single cut corner)



- ♦ Cut corner: A24
- ♦ Compatible with most PCR instruments
- ♦ Available with low adsorption and customizable barcoding



Length at base plane	127.76 mm
Width at base plane	85.48 mm
Height overall	9.6 mm
Well spacing	4.5 mm
Well volume	40 $\mu$ L
Well diameter at opening	4 mm
Cut corner	A24

### Ordering information

Part No.	Description	Package
PCRP-40-384S-C2	40 $\mu$ L 384-well single cut corner, black print, clear base, clear well	10 pcs/box, 10 boxes/case
PCRP-40-384S-W1	40 $\mu$ L 384-well single cut corner, black print, white base, clear well	10 pcs/box, 10 boxes/case

### Recommended Instrument Compatibility (Continuously Updated)

#### BIO-RAD:

C1000, C1000 Touch, S1000, CFX Opus 384, CFX384, CFX384 Touch, DNA Engine, Dyad Tetrad, Tetrad 2, PTC Tempo 384

#### ABI:

7300, 7500, ViiA7, 7500 Fast, ViiA7 Fast, 7900HT, 7900HT Fast, QuantStudio Systems, StepOne, StepOnePlus, ViiA 7 Plus

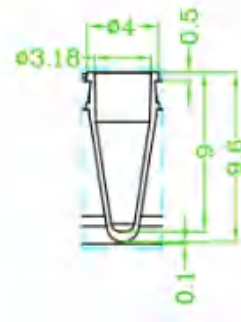
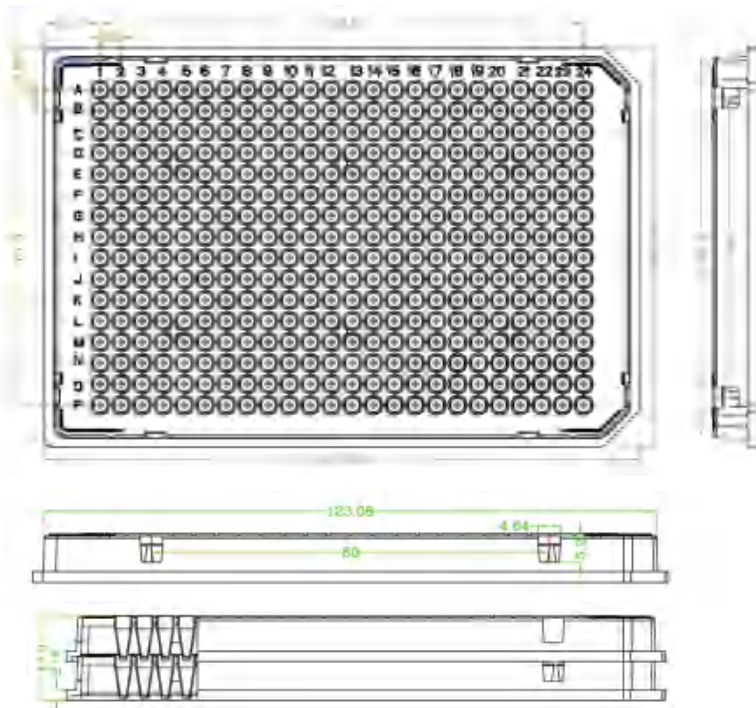
#### Eppendorf:

MasterCycler Series

## 40 $\mu$ L 384-well PCR plate (double cut corner)



- ◆ Cut corner: A24/P24
- ◆ Compatible with most PCR instruments
- ◆ Available with low adsorption and customizable barcoding



Length at base plane	127.76 mm
Width at base plane	85.48 mm
Height overall	9.6 mm
Well spacing	4.5 mm
Well volume	40 $\mu$ L
Well diameter at opening	4mm
Cut corner	A24/P24

### Ordering information

Part No.	Description	Package
PCRP-40-384D-C2	40 $\mu$ L 384-well double cut corner, black print, clear base, clear well	10 pcs/box, 10 boxes/case
PCRP-40-384D-C1	40 $\mu$ L 384-well double cut corner, black print, clear base, white well	10 pcs/box, 10 boxes/case
PCRP-40-384D-W2	40 $\mu$ L 384-well double cut corner, black print, white base, white well	10 pcs/box, 10 boxes/case

### Recommended Instrument Compatibility (Continuously Updated)

#### BIO-RAD:

CT, 1000™ Touch™, DNA Engine Dyad®/Dyad Disciple™, Engine Tetrad®2

## Five Things to Consider Before Buying Your Next PCR Plate

### Have you ever considered how your PCR plate could impact your results?

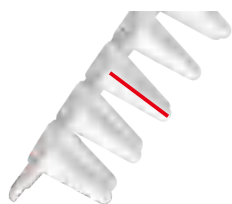
While often overlooked during experimental setup, PCR plates are just as critical to experimental success as the reagents and instruments themselves. Before initiating your next PCR run, consider these five key factors to ensure optimal performance.

#### 01 How Tall Should Your Plate Be?

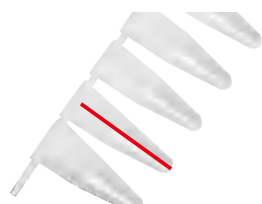
Have you ever pulled your PCR plate from your thermal cycler and noticed empty wells, condensation on the seal or cap, or crushed wells that bulge in the middle? These are all problems that can result if you use a PCR plate that is the wrong height for your thermal cycler.

The thermal lid assembly of PCR cyclers or real-time PCR instruments contains a heated compression plate mechanism. When the thermal lid closes, this mechanism applies specified pressure to the PCR plate. If using an over-height plate, it will cause excessive pressure, resulting in compression deformation of well walls during thermal cycling. Conversely, if using a low-profile plate in an instrument designed for high-profile plates, it will lead to insufficient pressure, which may consequently cause sample condensation or evaporation.

Instruments with adjustable lids are compatible with both high-and low-profile plates. One common identifying feature of these instruments is a wheel you can turn to tighten or loosen the lid. Some instruments automatically detect and adjust to different plate heights. These instruments give you the flexibility to select the best plate for your application. Alternatively, instruments with fixed-height lids can only be used with appropriately sized plates. Identifying your instrument's lid and plate specifications will help you to limit issues with your sealing method and avoid crushing wells.



Low-Profile



High-Profile

#### 02 What Color Wells Should Your Plate Have?

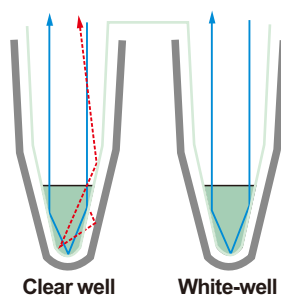
Did you know that the color of your PCR plate's wells can affect your real-time PCR results? PCR plates come in various well colors, the most common being clear or white. Each has distinct advantages and disadvantages which should be considered when choosing the right PCR plate for your experiment.

**Clear well plates** are constructed using polypropylene with no added colors and thus are translucent.

One advantage of clear wells is that they allow the user to visually monitor pipetting, as the sample volume can be observed from the bottom, sides, or top of the plate. Clear wells also allow you to see if a well has been missed or if volumes differ between wells.

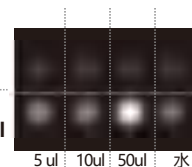
The main disadvantage of using clear-well plates is the possibility of fluorescence bleeding through. When the fluorescence signal from one well bleeds into a neighboring well, the results in the neighboring well will be artificially high. This situation is especially concerning when a sample with high fluorescence signal is adjacent to a sample with a low concentration of the target, as the bleed-through will artificially increase the signal coming from the low-concentration well.

**White well plates** are constructed using polypropylene with white color added to the resin, resulting in opaque wells. Seeing which wells contain samples and the volume of the sample in each well is more challenging in white-well plates because you can see the sample only when looking down into the well. Adding an inert dye to the sample helps minimize this drawback. White-well PCR plates offer a **significant advantage**: they prevent light from escaping through the sides of the opaque wells, increasing the signal-to-noise ratio. This better signal-to-noise ratio is critical when amplifying and detecting a signal from low-concentration samples. The opaque wells also eliminate any potential bleed-through into neighboring wells.



Clear well

White-well



5ul | 10ul | 50ul | 水

### 03 Does Your Plate Need a Skirt?

On a PCR plate, the skirt is the outer frame of the plate, providing plate rigidity and a flat surface for robotic plate handlers to grip. PCR plates have three different skirt options

**Unskirted plate** stops at the edge of the plate and does not have a vertical surface.

**Half-skirted plate** have a vertical surface that extends roughly halfway down the length of the wells.

**Full-skirted plate** has a vertical surface that extends the entire length of the wells and will rest on a bench.



Unskirted plate

Half-skirted plate

Full-skirted plate

The correct skirt length depends on your thermal cycler. Some cyclers encase the thermal block in the deck of the instrument, leaving no room for a skirt. Other instruments position the thermal block in open space, allowing for the use of half- and full-skirted plates. If using automation, the PCR plate must have a half or full skirt, which provides a surface for the robot to grip.

### 04 Does Your Plate Need a Rigid Frame?

PCR plates warping not only present handling challenges but may also compromise reaction efficiency. If a PCR plate does not sit flat in a thermal cycler block, the wells may not make consistent contact with the block, resulting in uneven heating of your samples. More critically, warping during thermal cycling may displace tube caps or optical seals, leading to sample evaporation. Such deformable plates are also incompatible with automated robotic handling systems.

PCR plates are produced in either one-component or two-component designs. One-component plates utilize pure polypropylene resin molded in a unitary construction. While polypropylene is particularly suitable for PCR applications-enabling thin-walled well formation and efficient thermal conduction during cycling-exclusive use of this material for both plate frame and wells presents inherent limitations. The polymer's inherent flexibility renders it susceptible to dimensional instability when exposed to repeated thermal cycling. This manifests as non-uniform polypropylene shrinkage, inducing plate warpage that compromises rim seal integrity. Subsequent seal failure permits sample evaporation, ultimately generating erroneous.

In contrast, PCR plates constructed using a two-component design are made with a polycarbonate frame and polypropylene wells. The frame is manufactured first, then the wells are molded into the frame. The advantage of using polycarbonate is that the frame can better withstand temperature changes, resulting in a rigid.

PCR plate less prone to warping. Since PCR plates constructed using a two-component design are rigid, the seal remains adhered to the plate, and samples are less likely to evaporate. This makes the two-component plate a much better choice for PCR assays. If you use a robotic plate handler to improve efficiency, accuracy, and throughput, you will want to use a plate constructed using a two-component design. Two-component plates are ideal for automation, as the rigid frame provides a surface for the robot to grip and hold, enabling accurate movement and alignment of the plate onto thermal cycler blocks.

### 05 Can You Trust Your PCR Plate Manufacturer?

Consistent performance across every plate and between production lots is critical for your PCR experiments. Maintaining rigorous quality control standards is fundamental to delivering reliably high-quality PCR plates.

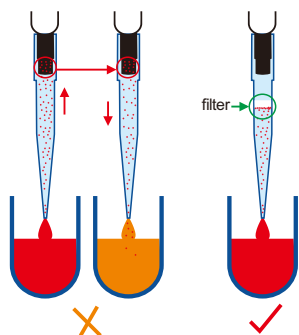
At PakGent, we implement stringent quality testing protocols throughout the entire manufacturing process to guarantee only the highest quality products reach your laboratory. Our Hard-frame PCR Plates are manufactured in our ISO 13485 & 9001 certified Centers of Excellence, where precision injection molding technology ensures exceptional product consistency and performance.

Our production processes are comprehensively monitored at every stage to guarantee the highest product quality, from raw material testing to in-process controls and final product inspections. Throughout manufacturing, we rigorously inspect our PCR plates to ensure dimensional consistency, minimal background fluorescence, and complete absence of defects. Additionally, each batch of PCR plates is accompanied by a Certificate of Analysis (COA) confirming the absence of DNase, RNase, and human DNA contamination.



## 9 Potential Real-Time PCR Pitfalls (and How to Avoid Them!)

### 01 Potential introduction of cross-contamination

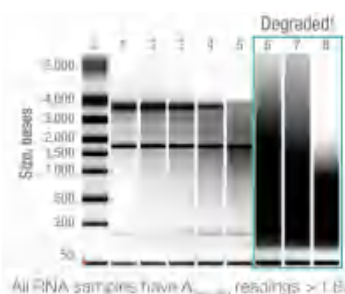


Amplified DNA is highly susceptible to aerosolization if proper handling precautions are not observed when manipulating reaction tubes. Contamination introduced into reaction vessels or reagents can significantly compromise data integrity, producing biologically irrelevant results.

#### How to avoid

- ♦ Use aerosol-resistant filter tips.
- ♦ Wear gloves and work in a dedicated aPCR area.
- ♦ Use screwcap tubes for template.
- ♦ Always use dedicated pipets for qPCR.
- ♦ Use PCR-grade water.
- ♦ Aliquot PCR components for single-time use.
- ♦ Clean bench with 10% bleach, not ethanol.
- ♦ Always include a no template control.

### 02 Using degraded RNA

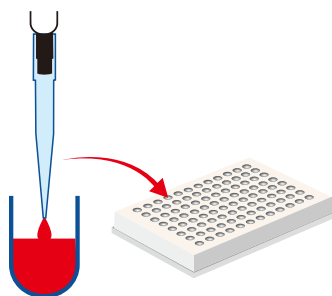


When it comes to qPCR, garbage in = garbage out. Degraded or contaminated RNA will yield low-quality cDNA, which will produce poor qPCR reaction efficiencies and result in low-quality data that are analytically inaccurate.

#### How to avoid

Assess the quality of your RNA by gel electrophoresis or by bioanalyzer analysis.

### 03 Pipetting reaction components separately



qPCR reaction efficiency is highly dependent on the chemical composition in the reaction vessel, thus each reaction must occur in an identical chemical environment. Pipetting components separately invites variability and error.

#### How to avoid

- ♦ Prepare a master mix that contains all reaction components except template, mix thoroughly, and dispense into each well.
- ♦ Prepare enough master mix to run all of your reactions plus 10% extra to compensate for potential pipetting errors. Do not freeze and reuse.

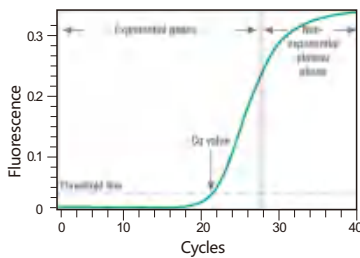
### 04 Forgetting to include controls

How can you verify proper target sequence amplification? Without controls, you cannot confirm assay specificity/accuracy or perform meaningful troubleshooting.

#### How to avoid

- ♦ **No template control:** Omit sample from 1 well to serve as a control for extraneous nucleic acid contamination.
- ♦ **No reverse transcription (RT) control:** For each assay include 1 well that uses a no reverse transcriptase cDNA sample as the template.
- ♦ **RNA quality control:** Use an RNA quality assay to verify RNA integrity.
- ♦ **Positive and negative controls:** Add a synthetic template to a reaction to demonstrate the reaction conditions are correct. Omit DNA polymerase from a negative control to assess background fluorescence signal.

## 05 Setting the threshold too high or too low



If the threshold is set outside the exponential growth phase of the reaction, the Cq value will not accurately represent the sample's DNA concentration, yielding biologically insignificant results.

### How to avoid

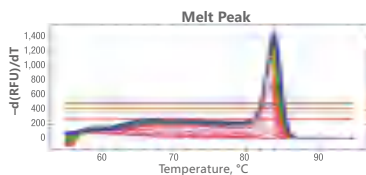
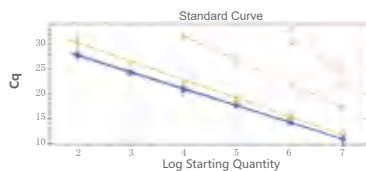
- If the plate includes serial dilutions, adjust the threshold to a position where you reach a maximum correlation coefficient ( $R^2$  value) for the standards.
- If the plate does not contain serial dilutions, place the threshold in the exponential growth phase, above the noise but below the plateau phase.

## 06 Failing to optimize and validate an assay

To achieve accurate template quantification in a qPCR assay, each reaction must efficiently amplify a single product. Amplification efficiency must be independent of:

- Template concentration.
- Amplification of other templates.
- Potentially contaminating compounds in the sample.

Even commercial assays should be validated under your specific and unique reaction conditions.



### How to avoid

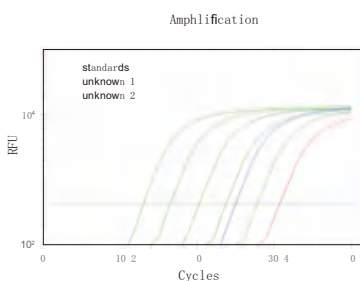
Always validate a new qPCR assay to verify its efficiency under your specific conditions.

- Determine efficiency using a standard curve spanning 5 orders of magnitude (5- or 10-fold dilutions) and run in triplicate to determine the efficiency, linear dynamic range, and reproducibility of the assay.
- Efficiency of the PCR should be 90-110%.
- $R^2$  of the standard curve should be  $>0.98$ .
- Cq values of replicates should vary by no more than 0.2 standard deviation units (Cq values).
- Identify the optimal annealing temperature by testing the amplification efficiency and reproducibility across a range of temperatures.
- Verify assay specificity by running a melt curve.

Temperature	Efficiency	$R^2$	Slope	Y-intercept
60.0°C	98.6%	1.000	3.357	34.294
60.7°C	97.1%	1.000	3.392	34.705
62.0°C	97.6%	0.999	3.381	34.529
64.0°C	95.6%	0.999	3.431	34.825
66.4°C	85.7%	0.995	3.719	37.715
68.4°C	61.3%	0.996	4.816	50.900
69.5°C	33.8%	0.872	7.898	79.112

## 07 Validating assay efficiency using an incorrect range of standard dilutions

The standard curve serves to evaluate three critical qPCR performance parameters: amplification efficiency, linear dynamic range, and assay reproducibility. However, these quantitative measurements remain valid solely within the concentration range represented by the serial dilution series. Importantly, reaction efficiency estimates cannot be reliably extrapolated to test samples exhibiting Cq values beyond the standard curve's empirically established Cq range.



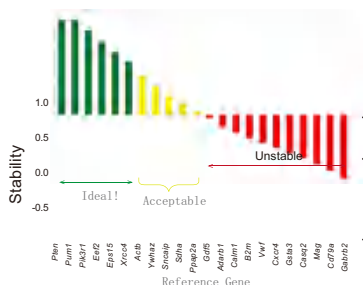
Unknown 2 is outside the linear range of the standard curve. Therefore, the PCR efficiency of the reaction is unknown for this sample.

### How to avoid

- Evaluate a range of standard dilutions that span the expected concentration range of your target.
- Prepare sequential dilutions (5- to 10-fold) that span at least 5 orders of magnitude.
- Pipet the same volume of DNA for each dilution.
- Use the correct size pipet, especially for small volumes.
- Avoid pipetting less than 5  $\mu$ L.
- Use water in place of DNA for a negative control to detect contaminants.

## 08 Using an unstable reference gene

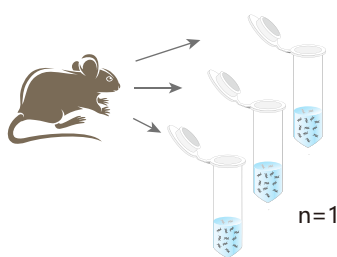
Not all commonly used reference genes are stable under all conditions. Failing to confirm the stability of a reference gene may produce biologically irrelevant results.



### How to avoid

- ◆ Validate the suitability of any reference gene to confirm its stability.
- ◆ Run a preplated reference panel containing a set of commonly used reference genes to identify the most suitable reference genes for your experimental condition.
- ◆ Use more than 1 reference gene that does not change expression as a result of the experimental treatment or condition.

## 09 Forgetting to use technical replicates



Reliable assessment of assay precision and reproducibility requires technical replicates of each sample, as single measurements cannot provide sufficient confidence in experimental outcomes.

### How to avoid

- ◆ Run each sample in triplicate (minimum).
- ◆ Perform a power analysis to determine how many replicates you need to see a certain fold change.

# PakGent®

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Address: No.9 Lecheng Road, Bixi Street, Changshu, Jiangsu Province, China.

**PakGent Bioscience (Shanghai) Co., Ltd.**  
Tel: 400-167-1717  
Address: Building C, No 888, Lake west road, Lin gang section, Free Trade Zone, Shanghai, China.