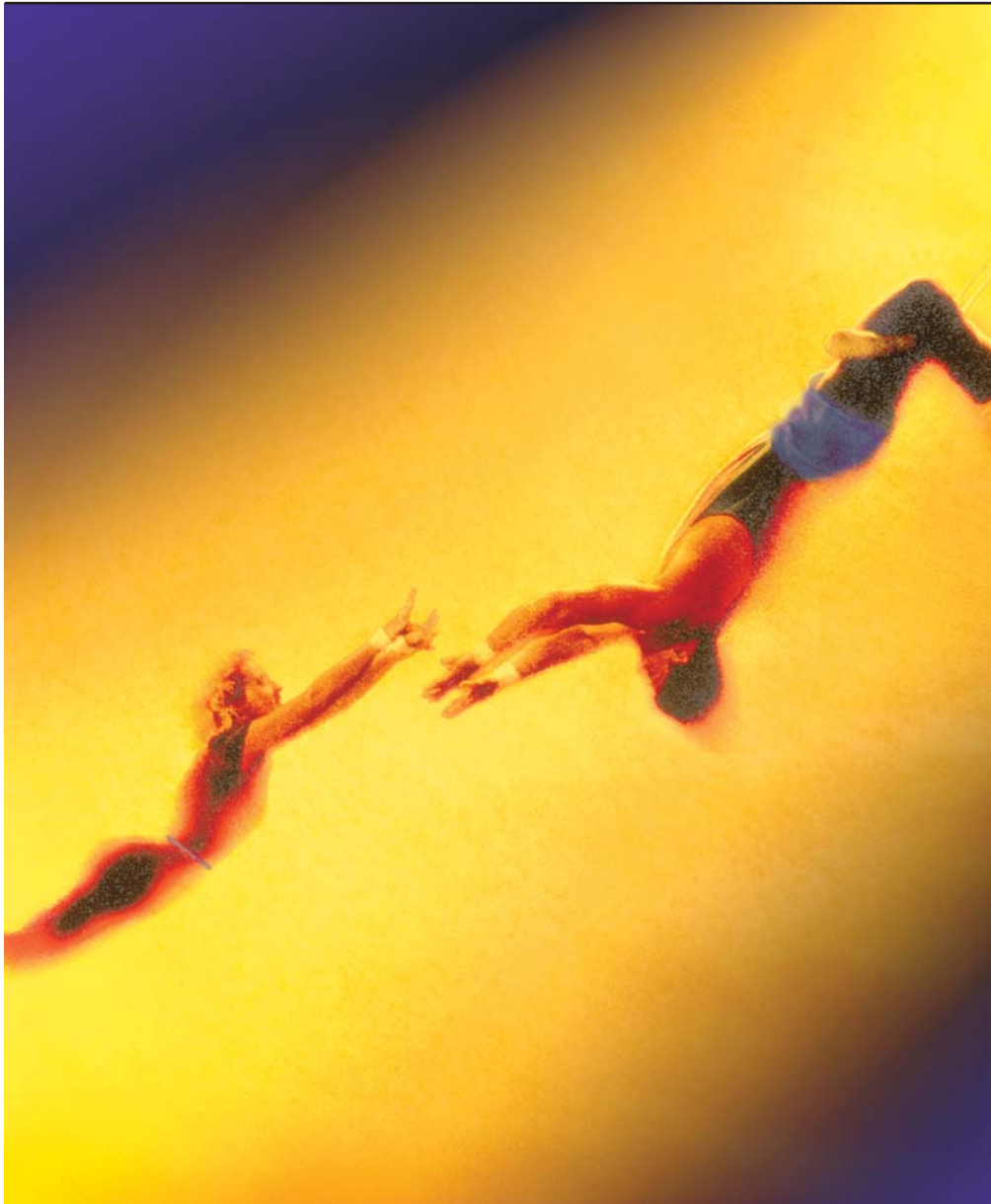




## Your Trusted Partner for Amplification



**Rely on the Invitrogen  
PCR and RT-PCR  
Resource for:**

- Higher specificity, fidelity, and performance
- Greater yield and sensitivity
- Amplification of long templates



For optimal amplification results, you need products you can rely on. Trust Invitrogen's PCR and RT-PCR technologies to deliver the highest yield and sensitivity, even with long or difficult templates. The Invitrogen PCR and RT-PCR resource offers the broadest array of products—DNA polymerases, reverse transcriptases, enzyme mixes, primers, and easy-to-use PCR and RT-PCR systems. You will find the best PCR and RT-PCR products to help you meet any challenge.

## Rely on Invitrogen

In your research you can't afford to take chances with unreliable products. You need amplification products that you can trust. Invitrogen's products and systems for PCR and RT-PCR will support you through every step of your research. You can count on these amplification products for the performance and reliability critical to your success.

Rely on Invitrogen to help you:

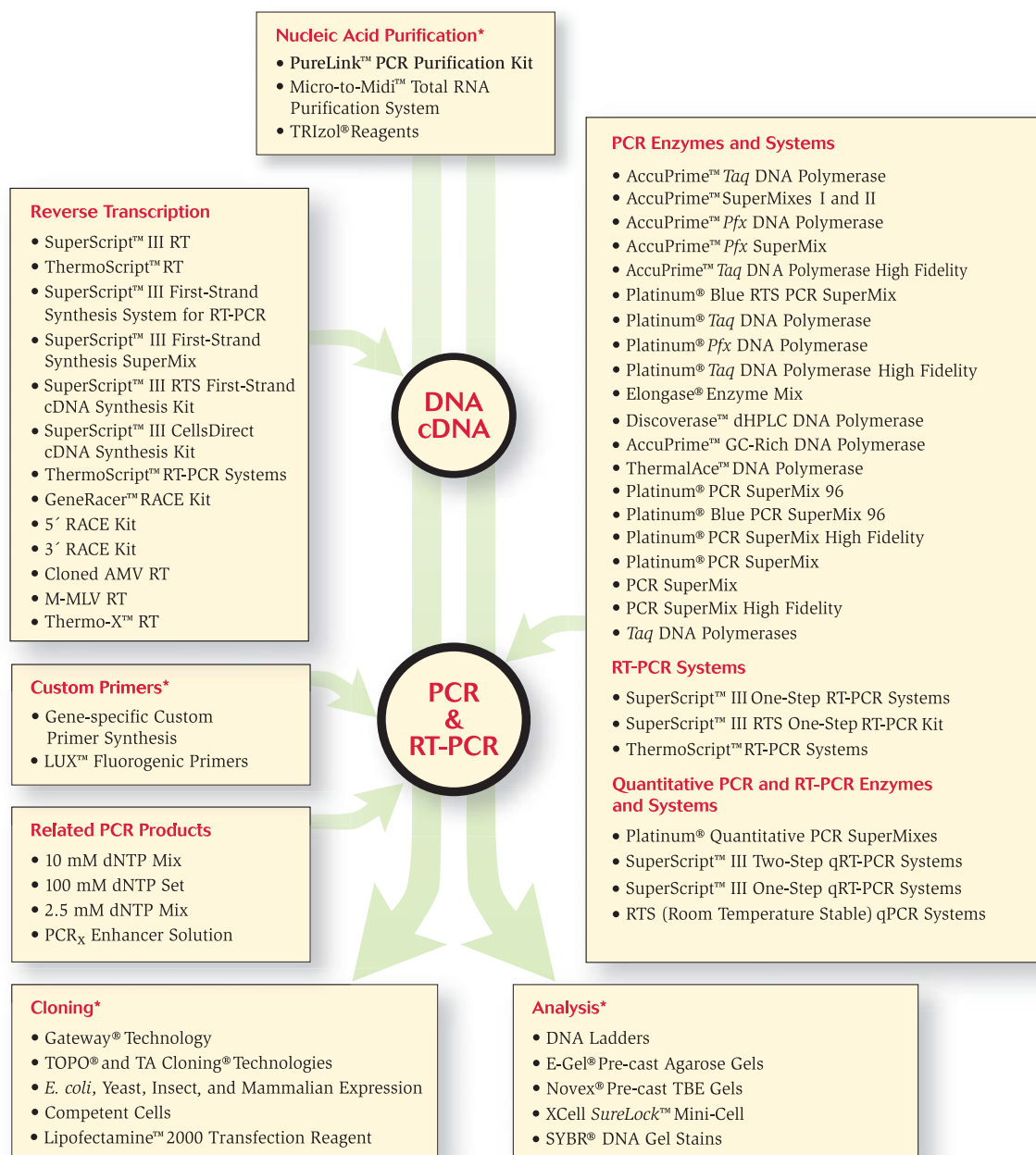
- Achieve higher specificity, fidelity, and performance
- Obtain greater yield and sensitivity
- Amplify longer templates
- Reduce time and eliminate steps

### Table of Contents

Description	Page
PCR Solutions	4
Routine PCR	5
Hot-Start PCR	6-7
High-Specificity PCR	8-9
High-Fidelity PCR	10-11
Robust PCR Enzyme Blends	12-14
High-Yield PCR for dHPLC	15
Problematic or GC-Rich PCR	16-17
Convenient PCR SuperMixes	18-19
Room Temperature Stable PCR SuperMixes	20
RT-PCR Solutions	21
Two-Step vs. One-Step RT-PCR	22
Highest-Yield Reverse Transcriptase	23
High-Throughput RT SuperMixes	24
Sensitive One-Step RT-PCR Kits	25
High-Fidelity RT-PCR	26
cDNA Directly From Cells	27
RTs for Difficult RNA Templates	29
Additional RT Products	30

# A comprehensive resource for each step of the process

No matter what your PCR or RT-PCR challenge is, you will find what you need to succeed with Invitrogen's PCR and RT-PCR resource. Use the chart below to find the right products for your specific application.



\* For more information on these products, please visit [www.invitrogen.com](http://www.invitrogen.com).

## PCR solutions

For increased yield, specificity, fidelity, and sensitivity when working with longer or more complex DNA targets, you need high-performance PCR enzymes. Invitrogen's Platinum® technology offers antibody-mediated hot-start PCR that reduces non-specific amplification to provide higher specificity, yield, and sensitivity than other DNA polymerases. While basic *Taq* polymerase provides

acceptable results for routine amplification, Platinum® technology is ideal for more complex PCR applications, such as gene expression analysis. For consistent, reproducible amplification, you can rely on Platinum® technology. In addition, AccuPrime™ accessory proteins provide superior priming accuracy and PCR specificity. See Tables 1 and 2 to determine the best product for your PCR application.

**Table 1 - PCR product details**

Invitrogen PCR Enzyme	Product Size	Yield	Specificity	Fidelity	Convenience	GC-rich
<i>Taq</i> DNA Polymerase	< 5 kb	•	•	•	•	•
PCR SuperMix	< 5 kb	•	•	•	•••	•
<i>Taq</i> PCR <sub>x</sub> DNA Polymerase	< 5 kb	•	•	•	••	•••
Platinum® <i>Taq</i> DNA Polymerase	< 5 kb	•••	••	•	••	•
Platinum® PCR SuperMix	< 5 kb	•••	••	•	••••	•
Platinum® PCR SuperMix 96	< 5 kb	•••	••	•	•••••	•
Platinum® Blue PCR SuperMix	< 5 kb	•••	••	•	•••••	•
Platinum® Blue RTS PCR SuperMix 96	< 5 kb	•••	••	•	••••••	•
Platinum® <i>Taq</i> PCR <sub>x</sub> DNA Polymerase	< 5 kb	•••	••	•	•••	•••
AccuPrime™ <i>Taq</i> DNA Polymerase	< 5 kb	•••	••••	••	•••	•
AccuPrime™ <i>Taq</i> SuperMix I and II	< 5 kb	•••	••••	••	••••	•
Platinum® <i>Pfx</i> DNA Polymerase	< 12 kb	••	••	•••••	•••	•
AccuPrime™ <i>Pfx</i> DNA Polymerase	< 12 kb	•••	•••	•••••	•••	•
AccuPrime™ <i>Pfx</i> SuperMix	< 12 kb	•••	•••	•••••	••••	•
Platinum® <i>Taq</i> DNA Polymerase High Fidelity	< 20 kb	••••	••	•••	••	•
Platinum® PCR SuperMix High Fidelity	< 20 kb	••••	••	•••	••••	•
AccuPrime™ <i>Taq</i> DNA Polymerase High Fidelity	< 20 kb	••••	•••	••••	•••	•
Elongase® Enzyme Mix	< 30 kb	•••	•	••	•	•
PCR SuperMix High Fidelity	< 12 kb	•••	•	••	•••	•
AccuPrime™ GC-Rich DNA Polymerase	< 5 kb	•••	•••	••	•	••••
ThermalAce™ DNA Polymerase	< 5 kb	•••	•	••	•	•••

\* For amplification of long, GC-rich templates, add PCR<sub>x</sub> Enhancer to either Platinum® *Taq* DNA Polymerase High Fidelity or Elongase® Enzyme Mix

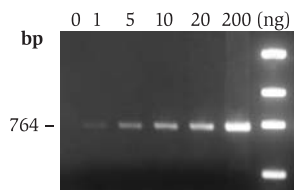
**Table 2 - Recommended products by application**

Application	PCR Target Length	Invitrogen Enzyme	Related PCR Enzyme Systems
<b>Routine PCR</b>	Up to 5 kb	<i>Taq</i> DNA Polymerase	PCR SuperMix, PCR Reagent System
<b>Automatic Hot-Start PCR</b>			
• Higher specificity	Up to 5 kb	Platinum® <i>Taq</i> DNA Polymerase	Platinum® PCR SuperMix
• Higher yield and sensitivity			Platinum® Blue PCR SuperMix
• Room temperature set-up			Platinum® Blue RTS PCR SuperMix 96
• Reduced primer-dimer artifacts	Up to 20 kb	Platinum® <i>Taq</i> DNA Polymerase High Fidelity	Platinum® PCR SuperMix High Fidelity
<b>High-Specificity PCR</b>			
• Highest specificity PCR	Up to 5 kb	AccuPrime™ <i>Taq</i> DNA Polymerase	PCR <sub>x</sub> Enhancer System
• Higher yield and sensitivity			AccuPrime™ SuperMix I and II
• No need for reaction optimization			
• Multiplexing			
• Higher accuracy (2X <i>Taq</i> )			
<b>High Fidelity PCR</b>			
• Accuracy 26X <i>Taq</i>	Up to 12 kb	Platinum® <i>Pfx</i> DNA Polymerase	
• Accuracy 6X <i>Taq</i>	Up to 20 kb	Platinum® <i>Taq</i> DNA Polymerase High Fidelity	
• Accuracy 5X <i>Taq</i>	Up to 30 kb	Elongase® Enzyme Mix	Add Platinum® Antibody for hot-start Platinum® PCR SuperMix High Fidelity
<b>High Fidelity plus High Specificity</b>			
• No need for reaction optimization			
• Accuracy 26X <i>Taq</i>	Up to 12 kb	AccuPrime™ <i>Pfx</i> DNA Polymerase	AccuPrime™ <i>Pfx</i> SuperMix
• Accuracy 9X <i>Taq</i>	Up to 20 kb	AccuPrime™ <i>Taq</i> DNA Polymerase High Fidelity	
<b>Problematic PCR</b>			
• High GC content templates	Up to 5 kb	AccuPrime™ GC-Rich DNA Polymerase	PCR <sub>x</sub> Enhancer System
• Codon repeats	Up to 5 kb	<i>Taq</i> PCR <sub>x</sub> DNA Polymerase	ThermalAce™ DNA Polymerase
	Up to 5 kb	Platinum® <i>Taq</i> PCR <sub>x</sub> DNA Polymerase	
	Up to 20 kb	PCR <sub>x</sub> Enhancer System plus Platinum® <i>Taq</i> DNA Polymerase High Fidelity	
	Up to 30 kb	Elongase® Enzyme Mix plus PCR <sub>x</sub> Enhancer System	

## Superior performance for your routine PCR

For performance you can depend on, choose Invitrogen's *Taq* DNA Polymerase. It offers higher yield and sensitivity, down to a single copy of genomic DNA (Figure 1), than other *Taq* polymerases on the market.

**Figure 1 - Amplification of single-copy gene from human genomic DNA**



Specific amplification of the single-copy gene coding for brain-derived neurotrophic factor (BDNF) from various amounts of human genomic DNA using *Taq* DNA Polymerase.

Product	Concentration	Quantity	Cat. no.
<b><i>Taq</i> DNA Polymerase</b>			
Native	5 units/ $\mu$ l	100 units	18038-018
	5 units/ $\mu$ l	500 units	18038-042
	5 units/ $\mu$ l	3 x 500 units	18038-067
	5 units/ $\mu$ l	5000 units	18038-240
<b><i>Taq</i> DNA Polymerase</b>			
Recombinant	5 units/ $\mu$ l	100 units	10342-053
	5 units/ $\mu$ l	500 units	10342-020
	5 units/ $\mu$ l	3 x 500 units	10342-046
	5 units/ $\mu$ l	5000 units	10342-178
PCR SuperMix		100 rxns	10572-014
<b>Nucleotides</b>			
10 mM dNTP Mix		100 $\mu$ l	18427-013
10 mM dNTP Mix		1 ml	18427-088
100 mM dNTP Set		4 x 25 $\mu$ mol	10297-018
100 mM dNTP Set		4 x 250 $\mu$ mol	10297-117
2.5 mM dNTP Mix		1 ml	R725-01
Custom Primers		visit <a href="http://www.invitrogen.com/pcr">www.invitrogen.com/pcr</a>	

## PCR Enzyme Selection Kits let you easily optimize your PCR results

It is often difficult to pinpoint the best PCR enzyme for a specific project. PCR Enzyme Selection Kits allow you to sample different Invitrogen PCR enzymes to choose the optimal enzyme for your template and experiment.

Two kit options, each containing four PCR enzymes, the corresponding buffers, and dNTPs, are available:

❖ The High Specificity Kit contains 50 reactions of each of the following enzymes:

- Platinum® *Taq* DNA Polymerase
- Platinum® *Taq* DNA Polymerase High Fidelity
- AccuPrime™ *Taq* DNA Polymerase
- Platinum® PCR SuperMix

❖ The High Fidelity Kit contains 50 reactions of each of the following enzymes:

- Platinum® *Pfx* DNA Polymerase
- AccuPrime™ *Pfx* DNA Polymerase
- Platinum® *Taq* DNA Polymerase High Fidelity
- Platinum® PCR SuperMix High Fidelity

Don't waste time optimizing sub-optimal enzymes; use the one that is best for your project right from the start.

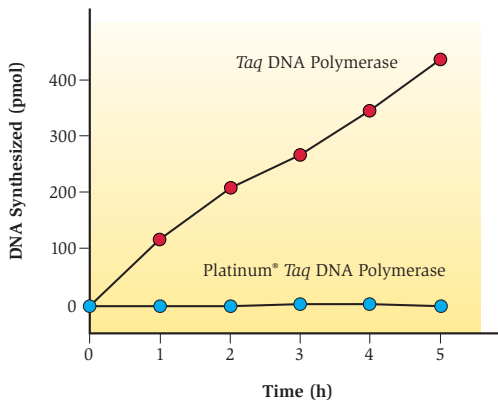
Product	Quantity	Cat. no.
PCR Enzyme Selection Kit		
- High Specificity	4 x 50 rxns	12567-012
PCR Enzyme Selection Kit		
- High Fidelity	4 x 50 rxns	12567-020

## Platinum® hot-start technology gives you higher specificity for higher yield

When you improve the specificity of your PCR reactions, you get greater yield of PCR product without non-specific amplification. With Platinum® automatic hot-start technology, proprietary antibodies inhibit enzyme activity at low temperatures (Figure 2). Up to 90% of *Taq* activity is restored in just minutes following antibody denaturation, unlike other hot-start technologies which

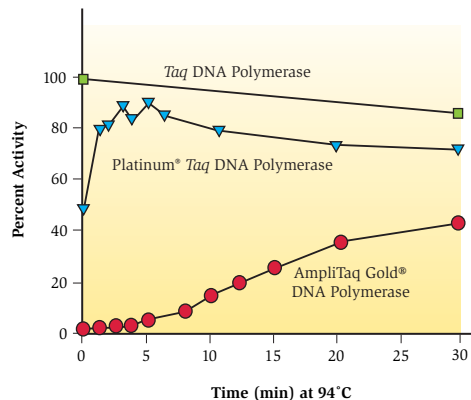
make modifications to the *Taq* that can hinder return to full activity (Figure 3). During the initial denaturation step, the antibodies are denatured and the enzyme is released into the reaction. In just minutes you have a fully active DNA polymerase. You'll get higher yields (Figure 4) and fewer extraneous bands due to non-specific amplification at lower temperatures (Figure 5, next page).

**Figure 2 - Complete inhibition of DNA polymerase activity for reaction set-up**



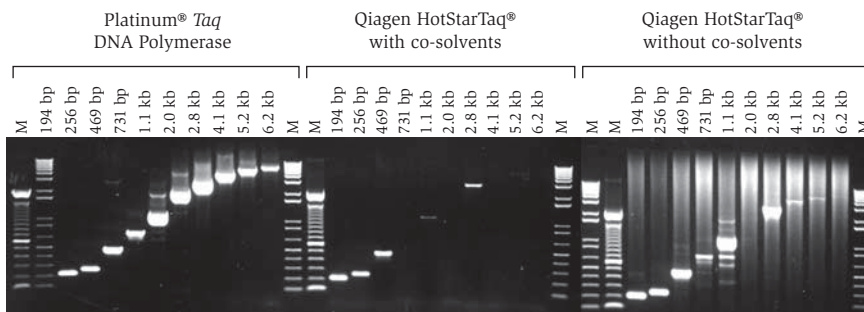
Activity assays were run at 37°C for 5 hours with *Taq* DNA polymerase and Platinum® *Taq* DNA Polymerase.

**Figure 3 - Enzyme recovery following thermal activation—Platinum® *Taq* compared to other DNA polymerases**



Enzymes were incubated at 94°C in the reaction buffer provided. Aliquots were removed at the various times and utilized in a standard activity assay.

**Figure 4 - Platinum® technology provides higher yield than other hot-start enzymes**



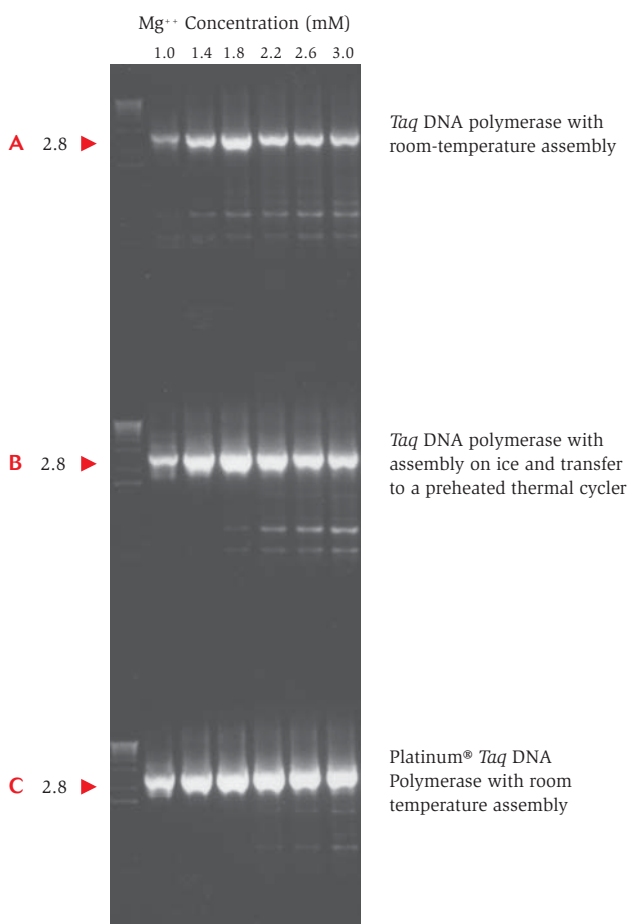
100 ng of K562 genomic DNA was amplified according to manufacturers' recommendations. Each reaction used 1.25 units of enzyme, 1X PCR buffer, 1.5 mM MgCl<sub>2</sub> and, 200 nM each primer, and was assembled at room temperature. Platinum® *Taq* was preincubated for 1 min. at 94°C and Qiagen HotStarTaq® was preincubated for 15 min. at 94°C.

### Higher sensitivity with limited template or less enzyme

Get higher yields of your desired product and improve the sensitivity of your PCR with Platinum® antibody-mediated hot-start technology. This is a great advantage when detecting expression of rare messages from a low concentration of target or working with a limited amount of starting material. With Platinum® hot-start

technology, you can also use fewer units of enzyme and still get better results than with other DNA polymerases (Figure 6). You'll use less enzyme per reaction, save money, and get higher sensitivity than with other hot-start enzymes.

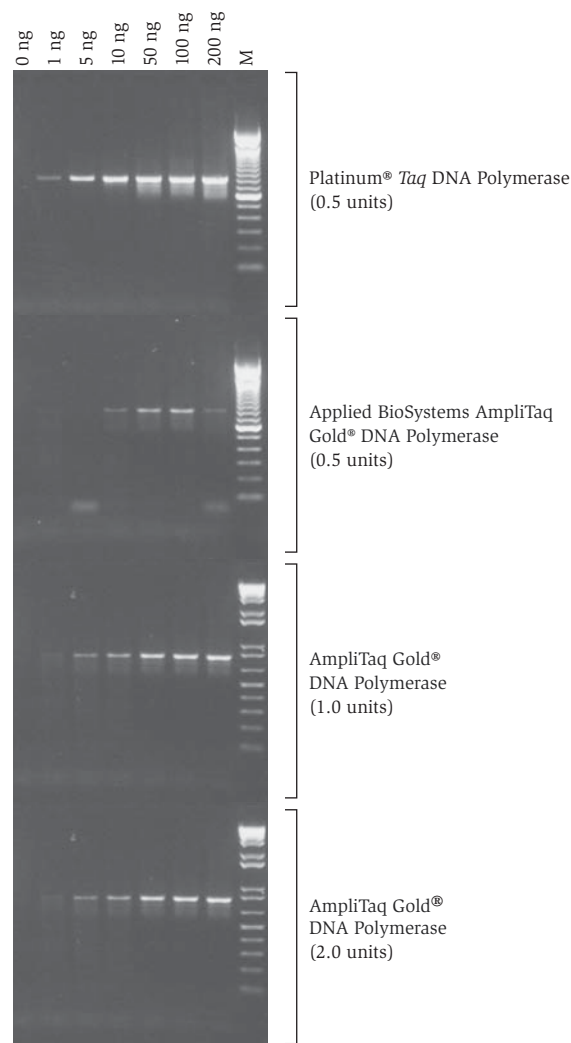
**Figure 5 - Reduce extraneous bands without re-optimization of existing protocols**



Amplification of a 2.8-kb region of the human  $\beta$ -globin gene using various  $MgCl_2$  concentrations.

Product	Quantity	Cat. no.	
Platinum® <i>Taq</i> DNA Polymerase	100 rxns	10966-018	
	250 rxns	10966-026	
	500 rxns	10966-034	
	5000 rxns	10966-083	
Platinum® PCR SuperMix	100 rxns	11306-016	
Platinum® PCR SuperMix 96	5 x 96-well	skirted	11306-065
		non-skirted	11306-073

**Figure 6 - Use up to four times less enzyme with Platinum® technology**



Human genomic DNA was amplified with Platinum® *Taq* and AmpliTaq Gold® according to manufacturers' recommendations. Platinum® *Taq* was preincubated for 1 minute at 94°C and AmpliTaq Gold® was preincubated for 10 minutes.

## AccuPrime™ *Taq* for the highest PCR specificity

AccuPrime™ *Taq* DNA Polymerase provides superior priming accuracy and PCR specificity that can't be achieved with other automatic hot-start enzymes. Anti-*Taq* DNA polymerase antibodies inhibit polymerase activity, providing an automatic hot-start. A proprietary AccuPrime™ accessory protein provides additional control over mispriming by interacting with template and primers to facilitate primer

binding to only the specific template sequence (Figure 7). While the antibodies are denatured in the first heating cycle of PCR, the accessory protein remains active throughout all PCR cycles to control mispriming. With AccuPrime™ *Taq*, you'll get extension of only the correct, specific PCR product (Figure 8).

Figure 7 - How AccuPrime™ *Taq* works

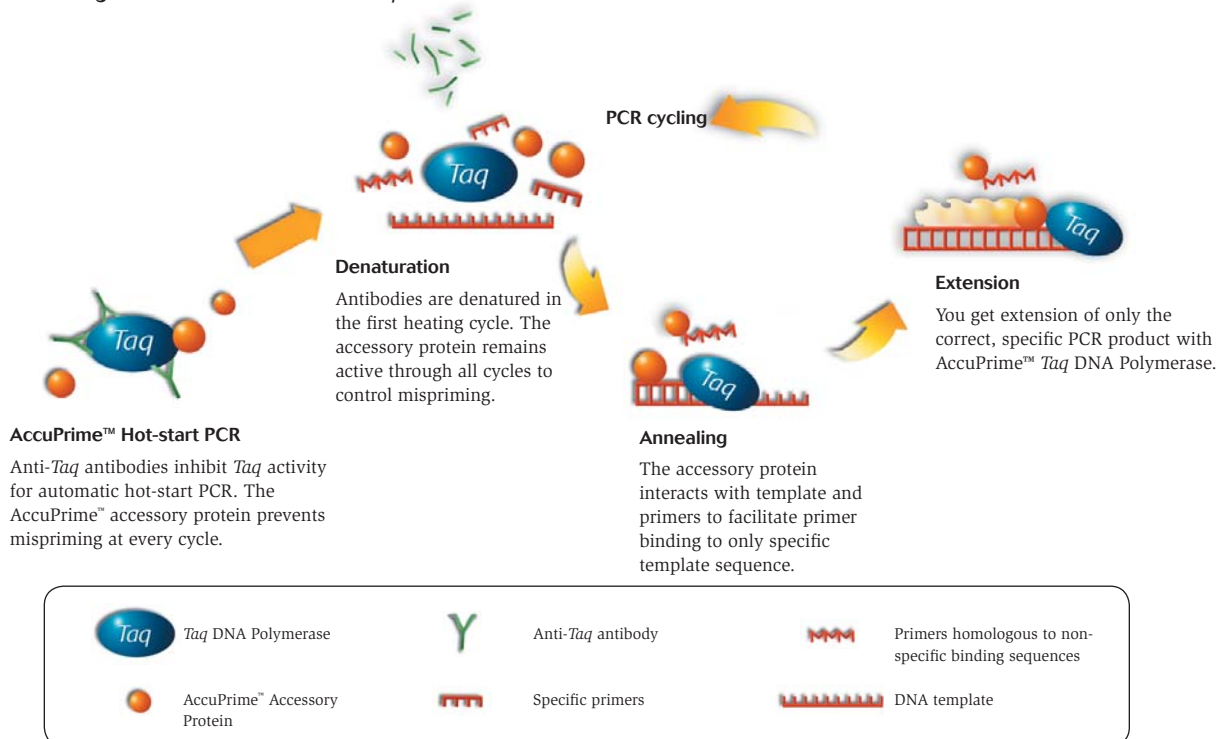
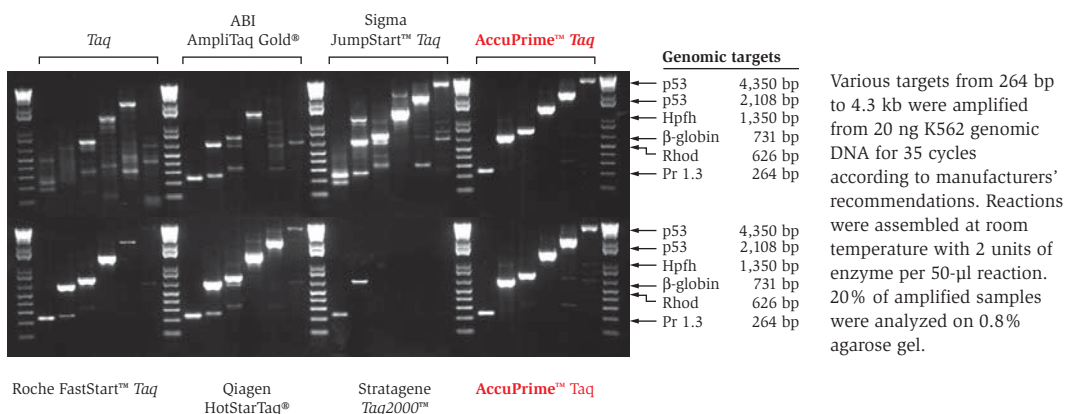


Figure 8 - High specificity with AccuPrime™ *Taq* DNA Polymerase



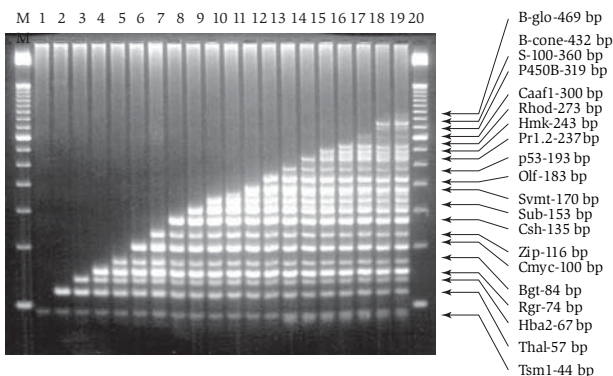


### High performance in multiplex and miniaturization

AccuPrime™ *Taq* DNA Polymerase is ideal for demanding PCR applications where the most robust PCR is required. In multiplex PCR, AccuPrime™ *Taq* delivers a single band for as many as 20 targets per reaction (Figure 9). And AccuPrime™ *Taq* controls non-specific amplification in miniaturized reactions down to 10 µl. With troublesome or poorly designed primer sets, or for reduction of

primer-dimers in PCR, AccuPrime™ *Taq* broadens primer annealing temperatures with robust performance between 55°C and 65°C. Primer design is less critical than with conventional hot-start enzymes, allowing you to overcome problems caused by sub-optimally designed primer sets. Don't waste your time and resources on redesigning primer sets; try AccuPrime™ *Taq*.

**Figure 9 - Resolution of up to 20 specific PCR products from a single-tube multiplex reaction**  
Progressive # of primer sets



Each lane from left to right represents the progressive number of primer sets (1-20) included in a single-tube 50-µl PCR reaction. PCR reactions were assembled on ice, using 200 ng K562 human genomic DNA, and five units AccuPrime™ *Taq* DNA Polymerase, and amplified for 35 cycles (94°C for 15 seconds, 60°C for 30 seconds, 68°C for one minute).

### Versatile technology

AccuPrime™ technology is available in a variety of configurations depending upon your needs. AccuPrime™ SuperMix I is designed for small genomic DNA amplicons (≤ 200 bp), plasmid DNA, or cDNA templates. AccuPrime™ SuperMix II is designed for genomic DNA

(200 bp-5 kb). AccuPrime™ *Taq* DNA Polymerase includes all the components needed for accurate amplification (except for template and primers), including both buffer systems (I and II).

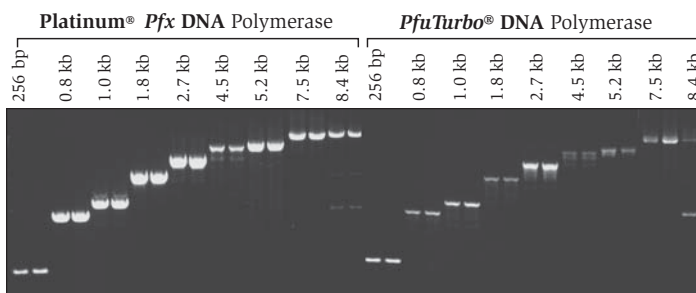
Product	Quantity	Cat. no.
AccuPrime™ <i>Taq</i> DNA Polymerase	200 rxns	12339-016
	1000 rxns	12339-024
AccuPrime™ SuperMix I	200 rxns	12342-010
	1000 rxns	12342-028
AccuPrime™ SuperMix II	200 rxns	12341-012
	1000 rxns	12341-020

## Highest fidelity PCR with Platinum® Pfx

When your PCR applications require high fidelity, choose Platinum® Pfx DNA Polymerase. This proprietary enzyme preparation containing recombinant DNA polymerase from *Thermococcus* species *KOD* offers all the features of Platinum® hot-start technology with the added benefit of proofreading activity. Proofreading enzymes possess 3'→5' exonuclease activity that enables the removal of

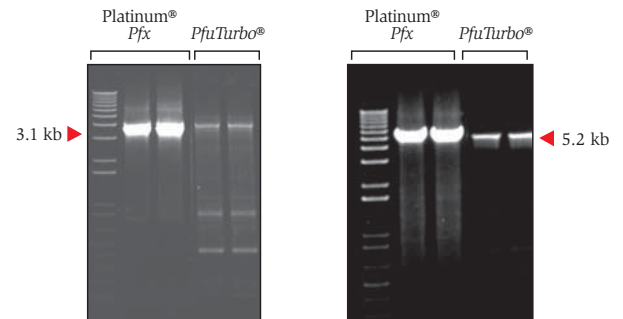
mismatched base pairings and decreases the error rate. While many commonly used proofreading enzymes are difficult to optimize and often generate low yields (Figure 10), Platinum® Pfx provides the high yield and fidelity you need without non-specific amplification (Figure 11).

**Figure 10 - Better yield over a wide range of target sizes**



200 ng K562 genomic DNA was amplified with 1.25 units of Platinum® Pfx DNA Polymerase or 2.5 units of Stratagene PfuTurbo®. Platinum® reactions were assembled at room temperature. PfuTurbo® DNA Polymerase reactions were assembled on ice.

**Figure 11 - High yield without the extra bands**

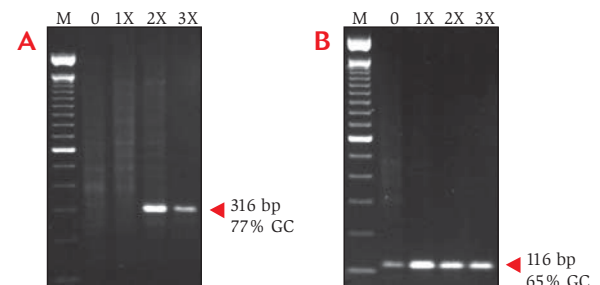


200 ng K562 genomic DNA was amplified with 1.25 units of Platinum® Pfx DNA Polymerase or 2.5 units of PfuTurbo®. Platinum® Pfx reactions were assembled at room temperature. PfuTurbo® reactions were assembled on ice.

### The lowest error rate even with problematic templates

When working with GC-rich sequences or other problematic templates where fidelity is a concern, Platinum® Pfx DNA Polymerase is the enzyme of choice. Platinum® Pfx is supplied with PCR<sub>x</sub> Enhancer Solution, a novel co-solvent that facilitates amplification of GC-rich sequences and problematic templates (Figure 12). The PCR<sub>x</sub> Enhancer Solution is also available separately for use with any Invitrogen™ DNA polymerase to optimize PCR of your difficult templates.

**Figure 12 - Amplification of GC-rich templates using PCR<sub>x</sub> Enhancer Solution**



Templates amplified using Platinum® Pfx DNA Polymerase and 0 to 3X PCR<sub>x</sub> Enhancer Solution. **Panel A:** Amplification of the CGG trinucleotide repeat region of the FRM-1 locus (Fragile X). **Panel B:** Amplification of a 116 bp, 65% GC-rich fragment of the AF064848 locus. M: 100 bp DNA ladder.

## Get the highest PCR fidelity and specificity with AccuPrime™ *Pfx*

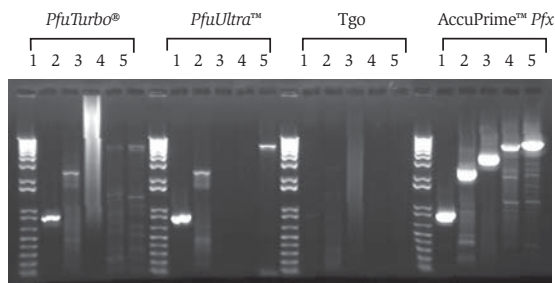
AccuPrime™ *Pfx* DNA Polymerase is the most accurate proofreading polymerase. It beats other proofreading enzymes in both specificity and yield (Figure 13).

### Formulated for precision

To enhance your PCR performance, AccuPrime™ *Pfx* DNA Polymerase incorporates:

- *Pfx* DNA Polymerase—thermostable proofreading enzyme with extremely low error rate
- Platinum® “hot start” anti-*Pfx* antibodies—to enhance specificity by blocking non-specific priming
- AccuPrime™ accessory proteins—to improve yields through prevention of mispriming during every PCR cycle

**Figure 13 - AccuPrime™ *Pfx* DNA Polymerase delivers higher yield and specificity than other proofreading enzymes**



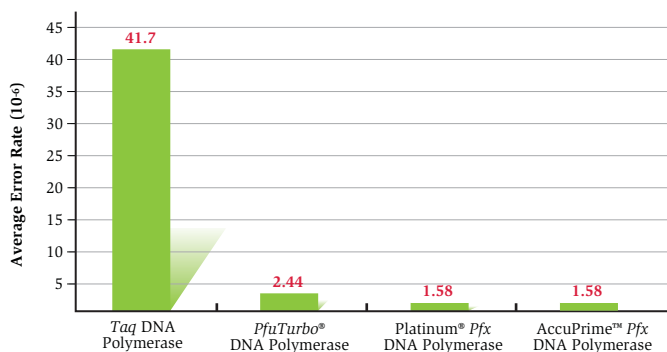
AccuPrime™ *Pfx* DNA Polymerase was tested side by side with three other proofreading enzymes. Each reaction was set up according to the enzyme manufacturer’s recommendations in 50-µl reaction volumes containing 1X buffer. 20% of each reaction was analyzed on a 0.8% agarose gel.

### Get fewer errors with Platinum® *Pfx* and AccuPrime™ *Pfx*

With Platinum® *Pfx* and AccuPrime™ *Pfx* DNA Polymerases you get:

- A lower error rate than other polymerases, including *PfuTurbo*® DNA Polymerase (Figure 14)
- Up to 20 times greater yield across a wide range of target sizes
- Cost savings by using only half the units required for other proofreading enzymes

**Figure 14 - Average error rates for DNA polymerases in the *rpsL* assay**



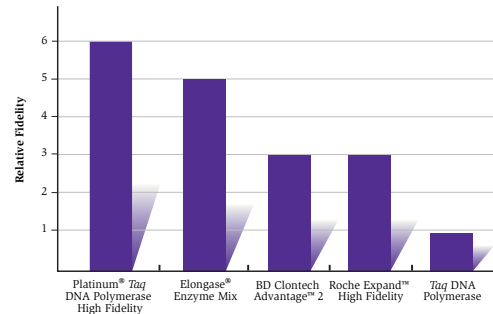
DNA was linearized and amplified with 2 units of *Taq* DNA Polymerase, 2.5 units of *PfuTurbo*® DNA Polymerase, 1.25 units of Platinum® *Pfx* DNA Polymerase, or 1 unit of AccuPrime™ *Pfx* DNA Polymerase.

Product	Quantity	Cat. no.
Platinum® <i>Pfx</i> DNA Polymerase	100 rxns	11708-013
	250 rxns	11708-021
	500 rxns	11708-039
AccuPrime™ <i>Pfx</i> DNA Polymerase	200 rxns	12344-024
	1000 rxns	12344-032
AccuPrime™ <i>Pfx</i> SuperMix	200 rxns	12344-040

## Balancing fidelity and yield, with the perfect blend

When yield is more important than fidelity, try Platinum® *Taq* DNA Polymerase High Fidelity or Elongase® Enzyme Mix. These products combine *Taq* with a proofreading enzyme for high fidelity with greater yield. Even though the addition of *Taq* results in lower fidelity than a proofreading enzyme alone, you'll get the best balance for your experiment. Try Platinum® *Taq* High Fidelity or Elongase® Enzyme Mix for higher fidelity and yield than *Taq* DNA polymerase alone (Figure 15). Both enzymes contain a mixture of *Taq* and proofreading *Pyrococcus* species *GB-D*, which provides 3'→5' exonuclease activity for increased fidelity. Platinum® *Taq* High Fidelity also includes anti-*Taq* antibodies for higher specificity and yield.

**Figure 15 - Fidelity of proofreading mixes versus *Taq* DNA polymerase**

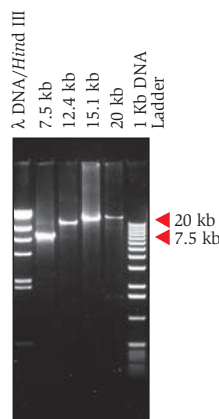


Relative fidelity of each enzyme is based on manufacturers' claims

## Elongase® Enzyme Mix for long PCR up to 30 kb

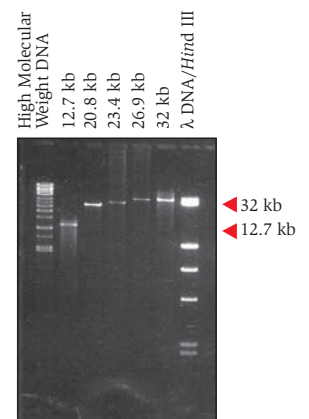
There is increasing demand for using PCR to amplify long templates. Although *Taq* has been used for targets as long as 5 kb, it offers a limited probability of success above 1 kb. For amplifying long PCR templates, try the Elongase® Amplification System or Enzyme Mix. You'll easily amplify genomic or λ DNA up to 30 kb (Figures 16A and B). Elongase® Enzyme Mix meets the challenge of even the longest PCR templates. If hot-start PCR is desired, combine Elongase® Enzyme Mix with the Platinum® *Taq* Antibody. Alternatively, you could use Platinum® *Taq* High Fidelity (page 13).

**Figure 16A - Amplification of long genomic DNA using Elongase® Enzyme Mix**



100 ng K562 human genomic DNA was amplified for 35 cycles. Magnesium concentration was optimized for each target.

**Figure 16B - Amplification of long λ DNA with Elongase® Enzyme Mix**



25 pg of λ DNA was amplified for 35 cycles. Magnesium concentration was optimized for each target.

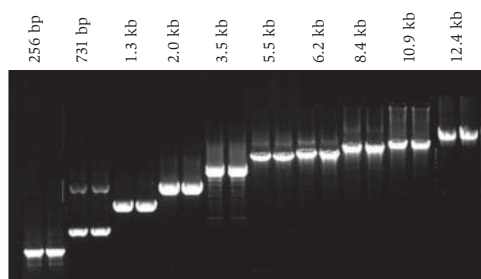
Product	Quantity	Cat. no.
Elongase® Enzyme Mix	100 rxns	10480-010
	500 rxns	10480-028
Elongase® Amplification System	100 rxns	10481-018
PCR SuperMix High Fidelity	100 rxns	10790-020

# Platinum® *Taq* DNA Polymerase High Fidelity provides the ideal balance

Use Platinum® *Taq* DNA Polymerase High Fidelity for amplifying templates from 100 bp to 20 kb. You'll amplify longer templates and still obtain the high sensitivity you expect from Platinum® hot-start technology (Figures 17A and B). You will

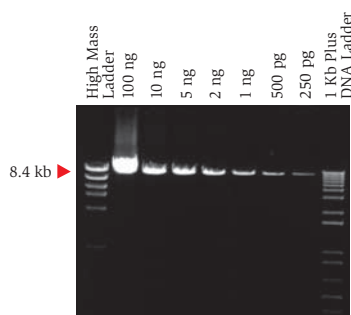
conserve precious starting material with detection as low as 1 pg, allowing you to detect rare messages that you may not see with other enzymes (Figure 18).

**Figure 17A - Amplification of increasing fragment lengths with Platinum® *Taq* High Fidelity**



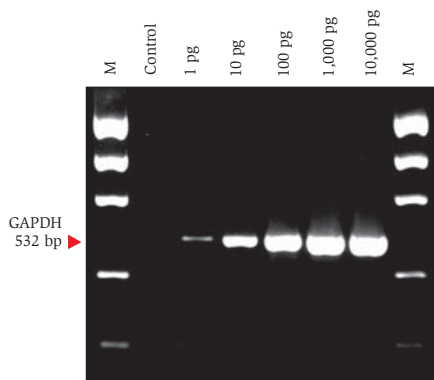
100 ng K562 human genomic DNA was amplified using 2.5 units of enzyme.

**Figure 17B - Sensitivity of long PCR with Platinum® *Taq* High Fidelity**



Varying amounts of K562 human genomic DNA were amplified using 2.5 units of enzyme.

**Figure 18 - Conserve starting material and detect rare messages**



GAPDH cDNA was synthesized from varying amounts of HeLa total RNA. One-tenth of the cDNA reaction was amplified with Platinum® *Taq* DNA Polymerase High Fidelity.

Product	Quantity	Cat. no.
Platinum® <i>Taq</i> DNA Polymerase High Fidelity	100 rxns	11304-011
	500 rxns	11304-029
	5000 rxns	11304-102
Platinum® PCR SuperMix High Fidelity	100 rxns	12532-016

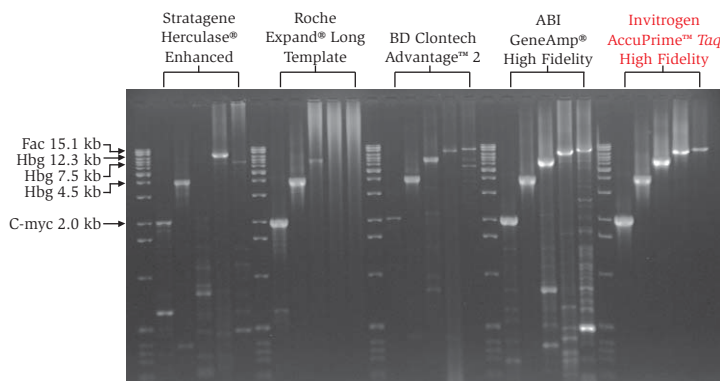
# AccuPrime™ *Taq* DNA Polymerase High Fidelity for improved specificity, yield, and robustness

AccuPrime™ *Taq* DNA Polymerase High Fidelity delivers all the advantages of Platinum® *Taq* DNA Polymerase High Fidelity plus AccuPrime™ accessory proteins to prevent mispriming during every cycle of PCR. The result is the most robust PCR enzyme blend available.

With AccuPrime™ *Taq* DNA Polymerase High Fidelity you get:

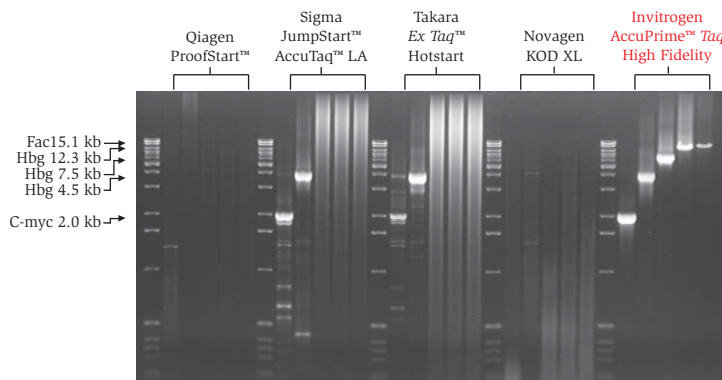
- The highest specificity and yield for the most robust PCR amplification (Figures 19A and B)
- 9-fold higher fidelity than *Taq* polymerase alone
- Minimal optimization steps, even with non-optimized primer sets
- Efficient amplification of targets over a broad size range up to 20 kb

**Figure 19A - AccuPrime™ *Taq* DNA Polymerase High Fidelity outperforms other enzyme blends**



Targets 2 kb through 15.1 kb were generated from 20 ng genomic DNA. Cycling conditions were performed as recommended by each manufacturer. All extensions were 1 minute per kb. 10% of each 50- $\mu$ l PCR reaction was loaded onto a 1% agarose gel.

**Figure 19B - AccuPrime™ *Taq* DNA Polymerase High Fidelity outperforms other enzyme blends**



Targets from 2 kb through 15.1 kb were generated from 20 ng genomic DNA. Cycling conditions were performed as recommended by each manufacturer. All extensions were 1 min. per kb. 10% of each 50- $\mu$ l PCR reaction was loaded onto a 1% agarose gel.

Product	Quantity	Cat. no.
AccuPrime™ <i>Taq</i> DNA Polymerase High Fidelity	200 rxns	12346-086
	1000 rxns	12346-094

## High-yield, high-fidelity PCR optimized for dHPLC applications

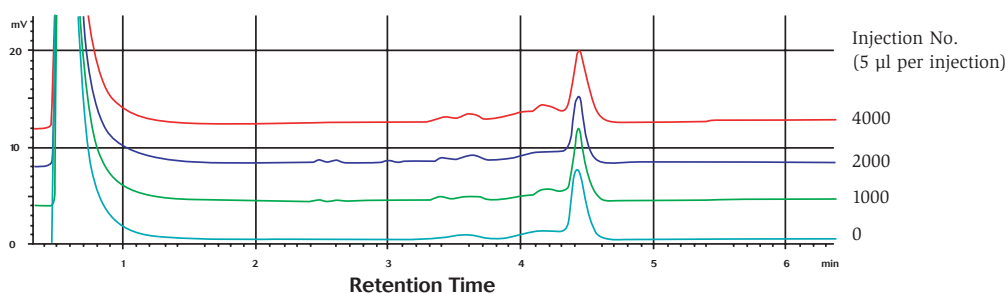
Discoverase™ dHPLC DNA Polymerase is a new high-fidelity PCR enzyme blend specifically designed for dHPLC and mutation analysis applications. Its high yield allows you to use less enzyme—saving you money.

### Compatibility with dHPLC

Discoverase™ dHPLC DNA Polymerase is optimized for mutation analysis instruments such as the Transgenomic WAVE® System. In accordance with dHPLC instrument manufacturers' recommendations, Discoverase™

Polymerase contains minimal detergents and will not interfere with the lifespan of dHPLC columns/cartridges (Figure 20).

Figure 20 - Discoverase™ dHPLC DNA Polymerase preserves the lifespan of dHPLC cartridges



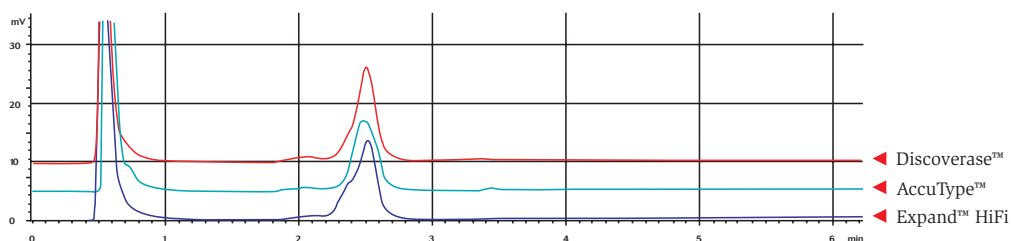
The retention time of PCR products generated with Discoverase™ dHPLC DNA Polymerase does not change over the course of 4000 5- $\mu$ l injections on the WAVE® System, demonstrating the compatibility of Discoverase™ with the DNASep™ Cartridge. To test this, eight hundred 25- $\mu$ l aliquots of Discoverase™ polymerase and buffer were injected onto the DNASep™ Cartridge and eluted using a linear acetonitrile gradient over 4.5 min at 58.6°C. A PCR product (rpsL542) was run after every 100 injections using a 5  $\mu$ l injection volume and the same gradient at 58.6°C. A cartridge active wash using 75% acetonitrile at 70°C was carried out after every 100 injections of reaction mix.

### High yield and high fidelity for robust PCR

To get the highest sensitivity from your dHPLC mutation detection applications, it is crucial to use an enzyme that offers high fidelity and robust amplification. Discoverase™ dHPLC DNA Polymerase is an enzyme blend of *Taq* DNA

polymerase and a proofreading enzyme. It is optimized for high-yield and high-fidelity amplification of targets < 1 kb (Figure 21) with 8-fold higher fidelity than *Taq* DNA polymerase alone.

Figure 21 - Discoverase™ dHPLC DNA Polymerase outperforms the competition



Discoverase™ dHPLC DNA Polymerase provides a higher PCR yield using significantly fewer units of enzyme per reaction. Using 100 ng of K562 human genomic DNA, target BRCA1 - 11F (416 bp) was amplified with 1 unit of Discoverase™, 2.5 units of AccuType™ Polymerase, and 2.6 units of Expand™ High Fidelity Polymerase, then subsequently analyzed on the WAVE® System.

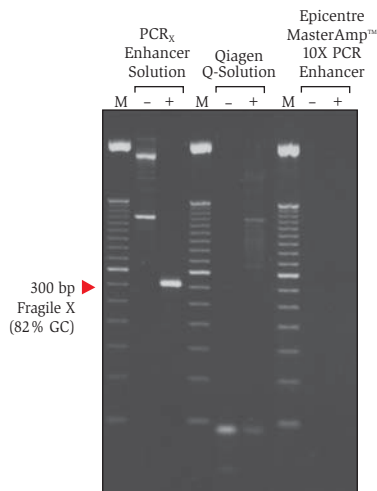
Product	Quantity	Cat. no.
Discoverase™ dHPLC DNA Polymerase	100 rxns	12607-016
	500 rxns	12607-024

## Optimize amplification of problematic templates

Templates with high GC content or codon repeats can be difficult to amplify. Several compounds have been found to help amplify these regions, but they also tend to inhibit polymerase activity. For improved yield and specificity, try the PCR<sub>x</sub> Enhancer Solution. Simply add PCR<sub>x</sub> Enhancer

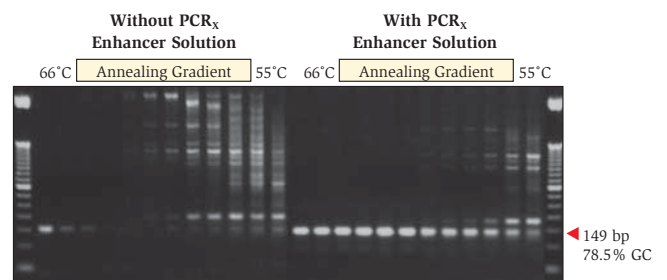
Solution and the amplification buffer to your reaction for higher yield and specificity than with other co-solvents (Figure 22). You'll also get easy optimization over a broader range of annealing temperatures and magnesium concentrations (Figures 23A and B).

**Figure 22 - High yield and specificity with PCR<sub>x</sub> Enhancer Solution**



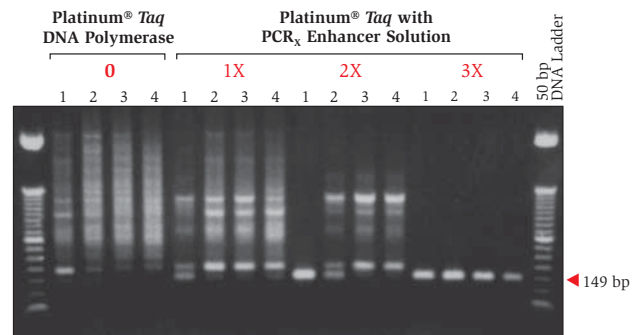
Amplification of the CGG trinucleotide repeat region of the FRM-1 locus (Fragile X) was performed using 1.25 units *Taq* DNA polymerase with PCR<sub>x</sub> Enhancer Solution and two other co-solvents according to their manufacturers' instructions.

**Figure 23A - Optimization over a broad range of annealing temperatures**



The 149-bp region of the AF064849 locus (78.5% GC) was amplified with Platinum<sup>®</sup> *Taq* DNA Polymerase in 1X PCR<sub>x</sub> Amplification Buffer without and with 1X PCR<sub>x</sub> Enhancer Solution.

**Figure 23B - Increased magnesium range with PCR<sub>x</sub> Enhancer Solution**



The 149-bp region of the AF064849 locus (78.5% GC) was amplified from human genomic DNA with Platinum<sup>®</sup> *Taq* DNA Polymerase in 1X PCR<sub>x</sub> Amplification Buffer with 0, 1X, 2X, or 3X PCR<sub>x</sub> Enhancer Solution. MgSO<sub>4</sub> concentration was 1.0, 1.5, 2.0, or 2.5 mM (lanes 1-4, respectively).

Product	Quantity	Cat. no.
PCR <sub>x</sub> Enhancer System	250 rxns	11495-017
<i>Taq</i> PCR <sub>x</sub> DNA Polymerase	500 units	11508-017
Platinum <sup>®</sup> <i>Taq</i> PCR <sub>x</sub> DNA Polymerase	500 rxns	11509-015

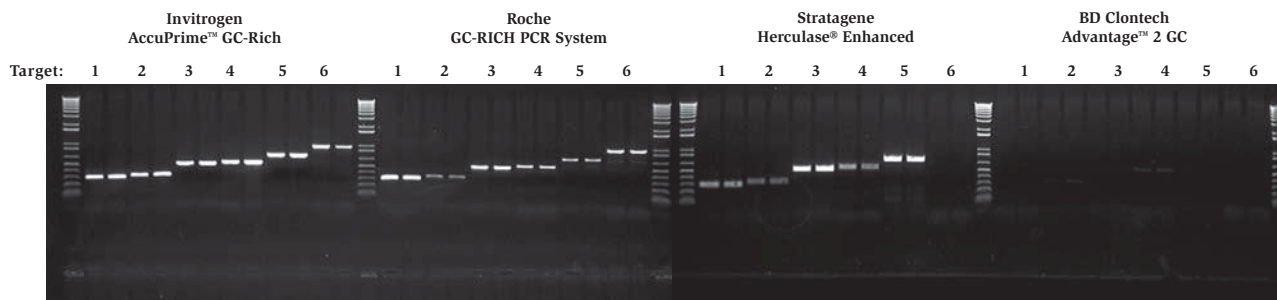


## Robust PCR for GC-rich templates

AccuPrime™ GC-Rich DNA Polymerase is optimized for difficult-to-amplify templates such as those containing > 65% GC content. AccuPrime™ GC-Rich outperforms the competition in robustness and yield (Figure 24).

In addition, AccuPrime™ GC-Rich DNA Polymerase is provided with two buffers to improve its flexibility— Buffer A is for genomic DNA targets, while Buffer B is for cDNA and plasmid targets.

Figure 24 - AccuPrime™ GC-Rich DNA Polymerase outperforms the competition



AccuPrime™ GC-Rich DNA Polymerase (in Buffer A) was compared to Roche’s GC-RICH PCR System, Stratagene’s Herculanse® Enhanced, and BD Clontech’s Advantage™ 2 GC using 100 ng K562 DNA with manufacturers’ recommended protocols and cycling conditions. The GC-rich targets used were 1) 299 bp, 2) 325 bp, 3) 520 bp 4) 545 bp 5) 709 bp, 6) 953 bp.

### High-specificity PCR

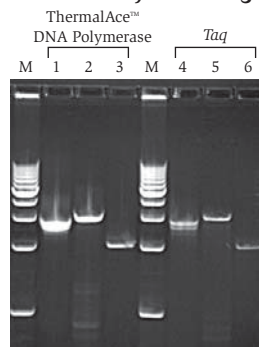
AccuPrime™ GC-Rich DNA Polymerase contains AccuPrime™ accessory proteins, which greatly improve the specificity and robustness of PCR. These AccuPrime™ proteins remain active throughout all PCR cycles,

facilitating primer binding to only the specific template sequence. This allows you to amplify only your specific DNA targets of interest, cycle after cycle.

## Great yields with ThermalAce™

ThermalAce™ DNA Polymerase provides high-fidelity, high-yield PCR amplification of a variety of templates (up to 5 kb). The key to the versatility of ThermalAce™, which was derived from a proprietary archea, is its extreme thermostability. This allows it to remain active for long periods at high temperatures (4 hours at > 95°C), dramatically improving PCR yields (Figure 25) and enhancing performance in amplification of problematic PCR products such as GC-rich templates.

Figure 25 - Relative yields using ThermalAce™ DNA Polymerase



Amplification of three separate templates using ten nanograms of HeLa first-strand cDNA for 30 cycles of PCR. Amplifications were performed using either ThermalAce™ or Taq DNA polymerase as indicated.

Lanes 1, 4: 3.0 kb thyroid hormone receptor coactivating gene  
 Lanes 2, 5: 3.3 kb Oncostatin-M specific receptor beta gene  
 Lanes 3, 6: 2.1 kb Portion of protein tyrosine Phosphatase Receptor beta gene

Product	Quantity	Cat. no.
AccuPrime™ GC-Rich DNA Polymerase	200 rxns	12337-016
	1000 rxns	12337-024
ThermalAce™ DNA Polymerase	200 units	E0200
	1000 units	E1000

## PCR SuperMixes save you time and lower the risk of contamination

PCR SuperMixes are ready-to-use mixtures that include all of the reagents needed for PCR in a convenient pre-mixed solution; just add your template and primers. By eliminating pipetting steps, you'll save time and reduce the risk of contamination (Figure 26).

Invitrogen PCR SuperMixes come in a variety of different enzyme compositions and formats so you can use them for any of your PCR applications:

**Platinum® PCR SuperMix** is a mixture of recombinant *Taq* DNA Polymerase, Platinum® hot-start antibodies, dNTPs, magnesium, and salts. As with Platinum® *Taq*, you'll get improved PCR specificity on targets up to 5 kb and the convenience of room temperature reaction setup, plus the added speed of a SuperMix format.

**Platinum® PCR SuperMix 96** is ideal for high-throughput applications. It offers the same benefits as standard Platinum® PCR SuperMix, in two convenient 96-well configurations. Choose either non-skirted, perforated plates or skirted plates to best suit your thermal cycler or robotic platform (Figure 27). The non-skirted plates offer perforations at 24-well increments to allow you to run as few as 24 or up to 96 reactions at a time. The unused wells can be stored at +4°C or -20°C until needed.

**Platinum® Blue PCR SuperMix** contains recombinant *Taq* DNA polymerase, Platinum® hot-start antibodies, dNTPs, magnesium, buffers, and an easy-to-track blue loading dye. All reagents are conveniently pre-mixed in one vial or pre-aliquoted into a 96-well plate, giving you the configuration to meet your research needs. Just add your template and primers and you're ready for PCR. After completing the PCR cycling, simply load your PCR product directly onto the agarose gel. That's it. No extra steps for mixing reagents or adding loading/tracking dye.

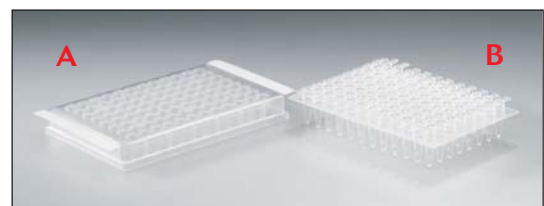
The blue loading/tracking dye in Platinum® Blue PCR SuperMix saves you time and effort, but does not inhibit your PCR performance. You'll achieve the same high yields as with the original Platinum® PCR SuperMix (Figure 28). Using Platinum® Blue PCR SuperMix, you'll efficiently amplify targets up to 5 kb.

**Figure 26 - Reaction set-up in half the time (for 24 reactions)**

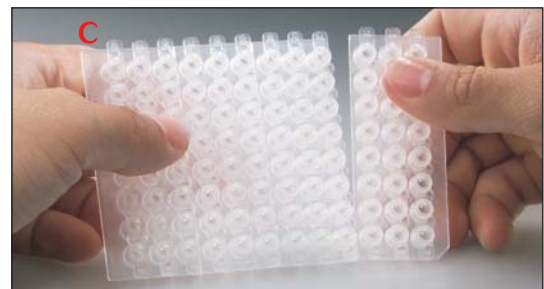
Individual Component Additions	PCR SuperMix Component Additions
Primer 1	Primer 1
+	+
Primer 2	Primer 2
+	+
DNA	DNA
+	+
Buffer	PCR SuperMix
+	
dNTPs	
+	
MgCl <sub>2</sub>	
+	
<i>Taq</i> DNA Polymerase	

Elapsed Time: 36 min      Elapsed Time: 18 min

**Figure 27 - Platinum® PCR SuperMix 96 plates**

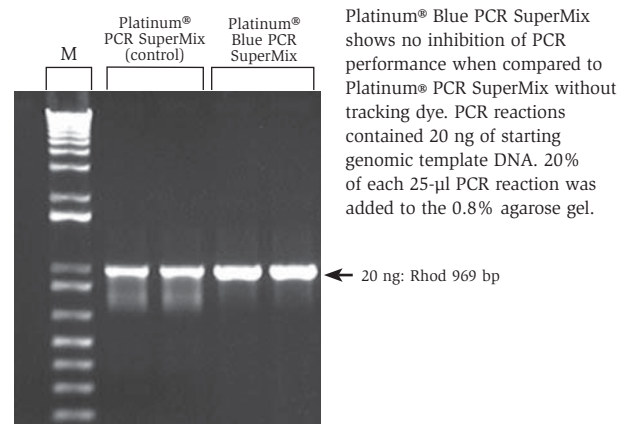


A: skirted plate B: non-skirted, perforated plate



C: non-skirted, perforated plates allow you to run only the desired number of reactions in 24-well increments

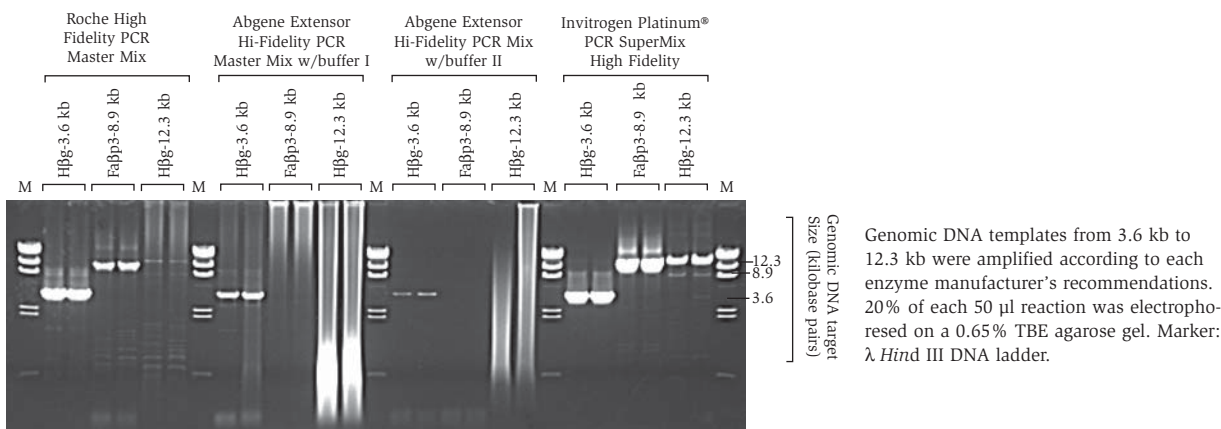
**Figure 28 - High PCR yield with Platinum® Blue PCR SuperMix**



**Platinum® PCR SuperMix High Fidelity** is a blend of Platinum® *Taq* DNA Polymerase and proofreading (3'→5' exonuclease activity) *Pyrococcus* species *GB-D* polymerase, dNTPs, magnesium, and salts. It utilizes Platinum® antibody-mediated hot-start technology for increased enzyme activity. Like Platinum® *Taq* DNA

Polymerase High Fidelity, this SuperMix gives you the perfect balance of high fidelity (6X higher than *Taq* DNA polymerase alone) and high product yields in your PCR with the convenience of a SuperMix format. Platinum® PCR SuperMix High Fidelity is ideal for templates up to 20 kb in length (Figure 29).

**Figure 29 - Platinum® PCR SuperMix High Fidelity outperforms other master mixes**



**PCR SuperMix High Fidelity** is designed for high-fidelity DNA amplification in applications such as cloning or mutagenesis, when you do not need the advantages of hot-start technology. It is a blend of recombinant *Taq* DNA Polymerase and proofreading (3'→5' exonuclease activity) *Pyrococcus* species *GB-D* polymerase, dNTPs, magnesium, and salts. You can amplify templates up to 12 kb in length.

**PCR SuperMix** is for routine PCR and provides efficient amplification of templates up to 5 kb. It combines the cost savings of *Taq* DNA Polymerase with the convenience of a SuperMix.

Choose the PCR SuperMix that best fits your research needs.

Product	Quantity	Cat. no.
Platinum® PCR SuperMix	100 rxns	11306-016
Platinum® PCR SuperMix 96 skirted	5 x 96-well plates	11306-065
Platinum® PCR SuperMix 96 non-skirted	5 x 96-well plates	11306-073
Platinum® Blue PCR SuperMix	100 rxns	12580-015
Platinum® Blue PCR SuperMix	1000 rxns	12580-023
Platinum® Blue PCR SuperMix 96 skirted	5 x 96-well	12580-031
Platinum® Blue PCR SuperMix 96 non-skirted	5 x 96-well	12580-049
Platinum® PCR SuperMix High Fidelity	100 rxns	12532-016
PCR SuperMix High Fidelity	100 rxns	10790-020
PCR SuperMix	100 rxns	10572-014

## Room temperature Stable (RTS) PCR SuperMixes

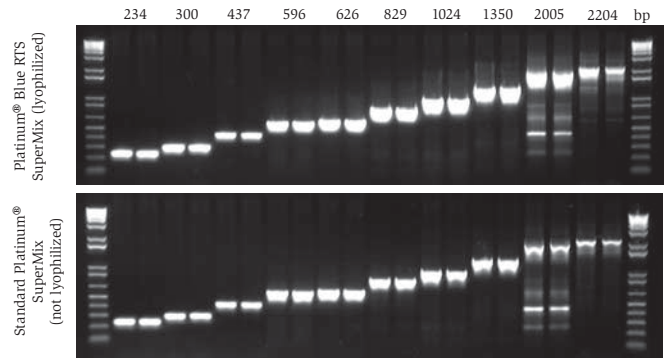
Platinum® Blue RTS PCR SuperMix combines easy-to-use reagents and industry-leading Platinum® *Taq* DNA Polymerase performance in one convenient, lyophilized format. A dried-down, single-use aliquot provides all the reagents you need for PCR amplification (including Platinum® *Taq* DNA Polymerase, Platinum® hot-start antibodies, dNTPs, loading dye, MgSO<sub>4</sub>, and buffer). There's no need to waste time thawing reagents. Simply add your primers and template to resuspend the reaction mixture, vortex briefly, and begin cycling. You'll get high yields and high specificity (Figure 30) with quicker reaction set up, fewer pipetting steps, and less chance of cross-contamination. RTS lyophilization technology ensures maximum enzyme stability for one year and high product yields with no ice requirements. The included pre-mixed blue loading dye lets you load samples directly onto a gel after thermal cycling, further reducing pipetting steps and allowing easy visualization of results.

### Formatted for your needs

Platinum® Blue RTS PCR SuperMix is available in skirted and semi-skirted 96-well plate configurations (Figure 31) to ensure compatibility with the most widely used instrument systems, while 12 x 8-strip wells in a plate format (Figure 32) accommodate smaller experiment setups. The semi-skirted plates are perforated in 24-well increments—just cut the plates at the perforations and use only the number of reactions you need. Store the remaining reactions at room temperature until needed.

Product	Quantity	Cat. no.
Platinum® Blue RTS PCR SuperMix 96 –		
skirted	5 x 96-well plates	12580-056
semi-skirted	5 x 96-well plates	12580-064
12 x 8-strip wells, plate format	12 x 8-strip wells	12580-072

**Figure 30 - Platinum® Blue RTS PCR SuperMix provides the same yield and specificity as standard Platinum® SuperMixes**



Agarose gel electrophoresis of 100 ng of K562 plasmid amplified with Platinum® Blue RTS PCR SuperMix (lyophilized, top gel) or standard Platinum® PCR SuperMix (unlyophilized, bottom gel). The performance of Platinum® Blue RTS PCR SuperMix was equal to that of the standard Platinum® PCR SuperMix in yield and specificity.

**Figure 31 - Platinum® Blue RTS PCR SuperMix skirted and semi-skirted 96-well plates**



**Figure 32 - Platinum® Blue RTS PCR SuperMix 12 x 8-strip well format**



## RT-PCR solutions

For successful RT-PCR you require reliable reverse transcriptases. Count on Invitrogen's reduced RNase H activity enzymes. SuperScript™ III RT prevents RNA degradation and provides greater thermostability and longer half-life for higher cDNA yields and greater success with gene-specific primers. Active at temperatures up to 80°C, Thermo-X™ RT allows you to

work through challenging RNA secondary structures, improving the efficiency of your cDNA synthesis. For the sensitivity and specificity you need with reduced RNase H activity, rely on SuperScript™ III RT and for the highest thermostability rely on Thermo-X™ RT. See Tables 4 and 5 to determine the best RT-PCR product for your application.

**Table 4 - RT-PCR product details**

Invitrogen RT Enzyme or RT-PCR System	Target Size	Sensitivity	PCR Fidelity	Convenience
M-MLV Reverse Transcriptase	≤ 12 kb	•	•	••
Cloned AMV Reverse Transcriptase	≤ 9 kb	•	•	••
SuperScript™ III Reverse Transcriptase	≤ 12 kb	•••	•	••
SuperScript™ III First-Strand Synthesis System for RT-PCR	≤ 12 kb	•••	•	•••
SuperScript™ III First-Strand Synthesis SuperMix	≤ 12 kb	•••	•	••••
SuperScript™ III RTS First-Strand cDNA Synthesis System	≤ 12 kb	•••	•	•••••
SuperScript™ III CellsDirect™ cDNA Synthesis Kit	≤ 4.5 kb	•••••	•	•••••
SuperScript™ III One-Step RT-PCR System with Platinum® Taq DNA Polymerase	≤ 4.5 kb	•••••	•	•••••
SuperScript™ III RTS One-Step RT-PCR System with Platinum® Taq	≤ 4.5 kb	•••••	•	••••••
SuperScript™ III One-Step RT-PCR System with Platinum® Taq High Fidelity	≤ 9 kb	•••••	••	•••••
Thermo-X™ Reverse Transcriptase	≤ 12 kb	•••	•	••
ThermoScript™ Reverse Transcriptase	≤ 12 kb	•••	•	••
ThermoScript™ RT-PCR System with Platinum® Taq DNA Polymerase	≤ 3.5 kb	•••	•	••••
ThermoScript™ RT-PCR System with Platinum® Taq DNA Polymerase High Fidelity	≤ 12 kb	•••	••	••••

**Table 5 - Recommended products by application**

Application	One-Step RT-PCR		Two Step RT-PCR	
			RT	PCR
<b>Higher Yield and Sensitivity</b> <ul style="list-style-type: none"> <li>• Low copy detection</li> <li>• Rare message</li> <li>• Limited sample quantity</li> <li>• More full-length cDNA</li> </ul>	<ul style="list-style-type: none"> <li>• SuperScript™ III One-Step RT-PCR System with Platinum® Taq</li> <li>• SuperScript™ III One-Step RT-PCR System with Platinum® Taq High Fidelity</li> </ul>	<ul style="list-style-type: none"> <li>• SuperScript™ III First-Strand Synthesis SuperMix</li> <li>• SuperScript™ III First-Strand Synthesis System for RT-PCR</li> </ul>	<ul style="list-style-type: none"> <li>• Platinum® Taq DNA Polymerase</li> <li>• Platinum® Taq DNA Polymerase High Fidelity</li> </ul>	
<b>Room Temperature Setup</b> <ul style="list-style-type: none"> <li>• Remote use</li> </ul>	<ul style="list-style-type: none"> <li>• SuperScript™ III RTS One-Step RT-PCR Kit with Platinum® Taq</li> </ul>	<ul style="list-style-type: none"> <li>• SuperScript™ III RTS First-Strand cDNA Synthesis Kit</li> </ul>	<ul style="list-style-type: none"> <li>• Platinum® Taq DNA Polymerase</li> <li>• Platinum® Taq DNA Polymerase High Fidelity</li> </ul>	
<b>Higher Yield from Cells</b> <ul style="list-style-type: none"> <li>• Single cell detection</li> <li>• Rare message</li> </ul>		<ul style="list-style-type: none"> <li>• SuperScript™ III CellsDirect™ cDNA Synthesis Kit</li> </ul>	<ul style="list-style-type: none"> <li>• Platinum® Taq DNA Polymerase</li> <li>• Platinum® Taq DNA Polymerase High Fidelity</li> </ul>	
<b>Problematic RNA</b> <ul style="list-style-type: none"> <li>• High secondary structure</li> <li>• GC-rich content</li> <li>• High Thermostability</li> </ul>		<ul style="list-style-type: none"> <li>• Thermo-X™ Reverse Transcriptase</li> <li>• ThermoScript™ Reverse Transcriptase</li> <li>• ThermoScript™ RT-PCR System with Platinum® Taq DNA</li> <li>• ThermoScript™ RT-PCR System with Platinum® Taq DNA</li> </ul>	<ul style="list-style-type: none"> <li>• Platinum® Taq DNA Polymerase</li> <li>• Platinum® Taq DNA Polymerase High Fidelity</li> </ul>	
<b>Higher Fidelity RT-PCR</b> <ul style="list-style-type: none"> <li>• High secondary structure</li> </ul>	<ul style="list-style-type: none"> <li>• SuperScript™ III One-Step RT-PCR System with Platinum® Taq High Fidelity</li> </ul>	<ul style="list-style-type: none"> <li>• SuperScript™ III First-Strand Synthesis SuperMix</li> <li>• SuperScript™ III First-Strand Synthesis System for RT-PCR</li> <li>• SuperScript™ III RTS First-Strand cDNA Synthesis Kit</li> </ul>	<ul style="list-style-type: none"> <li>• Platinum® Taq DNA Polymerase High Fidelity</li> <li>• Platinum® Pfx DNA Polymerase</li> </ul>	
<b>Routine RT-PCR</b> <ul style="list-style-type: none"> <li>• High copy detection</li> <li>• Abundant sample quantity</li> </ul>		<ul style="list-style-type: none"> <li>• Cloned AMV Reverse Transcriptase</li> <li>• M-MLV Reverse Transcriptase</li> </ul>	<ul style="list-style-type: none"> <li>• Platinum® Taq DNA Polymerase</li> </ul>	

## Two-Step vs. One-Step RT-PCR

### Two-Step

Two-Step RT-PCR is useful for detecting multiple messages from a single RNA sample. You'll get greater flexibility when choosing polymerase and primers than with one-step RT-PCR systems. When performing two-step RT-PCR you have the option of using either oligo(dT), random hexamers, or gene-specific primers, and then performing PCR in combination with either Platinum® *Taq* DNA Polymerase, Platinum® *Taq* DNA Polymerase High Fidelity, or your choice of other PCR enzymes.

### One-Step

One-Step RT-PCR allows easier processing of large numbers of samples, and helps minimize carry-over contamination since tubes are not opened between cDNA synthesis and amplification. By amplifying the entire cDNA sample, one-step RT-PCR can provide greater sensitivity-down to 0.01 pg total RNA. Table 6 compares the benefits of one-step and two-step RT-PCR procedures.

**Table 6 – Comparison of one-step and two-step RT-PCR procedures**

	Two-Step Procedure	One-Step Procedure
Prime first-strand cDNA with:	Oligo(dT) primer Random hexamers Gene-specific primers	Gene-specific primers
Provides:	Flexibility <ul style="list-style-type: none"> <li>• Choice of primer</li> <li>• Choice of amplification system</li> <li>• Ability to save some RNA sample for later use</li> <li>• Ability to optimize for difficult RT-PCR (combine with Platinum® enzymes for higher specificity or combine with Platinum® <i>Pfx</i> for greater fidelity)</li> </ul>	Convenience <ul style="list-style-type: none"> <li>• Amplification enzymes premixed with reverse transcriptase</li> <li>• Fewer pipetting steps and reduced chances of contamination</li> <li>• High sensitivity</li> </ul>
Recommended uses:	Ideal for detection or quantifying several messages from, a single sample	Ideal for analysis of large numbers of samples Ideal for real-time quantitative RT-PCR
Invitrogen Products	SuperScript™ III Reverse Transcriptase SuperScript™ III First-Strand Synthesis System for RT-PCR SuperScript™ III First-Strand SuperMix SuperScript™ III RTS First-Strand cDNA Synthesis Kit SuperScript™ III CellsDirect cDNA Synthesis Kit Thermo-X™ Reverse Transcriptase ThermoScript™ Reverse Transcriptase ThermoScript RT-PCR System for First-Strand cDNA Synthesis ThermoScript™ RT-PCR System and Platinum® <i>Taq</i> DNA Polymerase ThermoScript™ RT-PCR System and Platinum® <i>Taq</i> DNA Polymerase High Fidelity	SuperScript™ III One-Step RT-PCR System with Platinum® <i>Taq</i> DNA Polymerase SuperScript™ III One-Step RT-PCR System with Platinum® <i>Taq</i> High Fidelity SuperScript™ III RTS One-Step RT-PCR Kit

## Higher thermostability, longer half-life mean better performance with SuperScript™ III RT

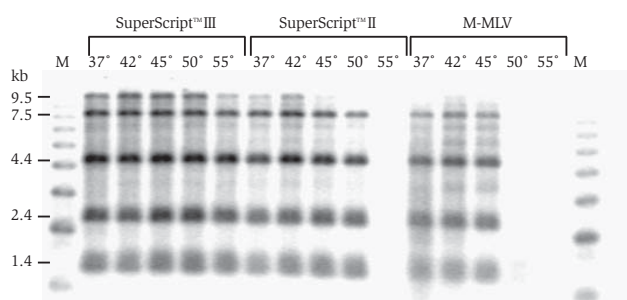
SuperScript™ III RT, a more thermostable mutant of SuperScript™ II RT, is simply the best-performing RT available for your gene expression studies. Genetically engineered to provide increased half-life, reduced RNase H activity, and increased thermal stability, SuperScript™ III gives you:

- A half-life of 220 minutes at 50°C, for the highest cDNA yields (Figure 33)
- Reduced RNase H activity for more full-length cDNA (Figure 34)

- Full activity at 50°C for increased specificity with gene-specific primers

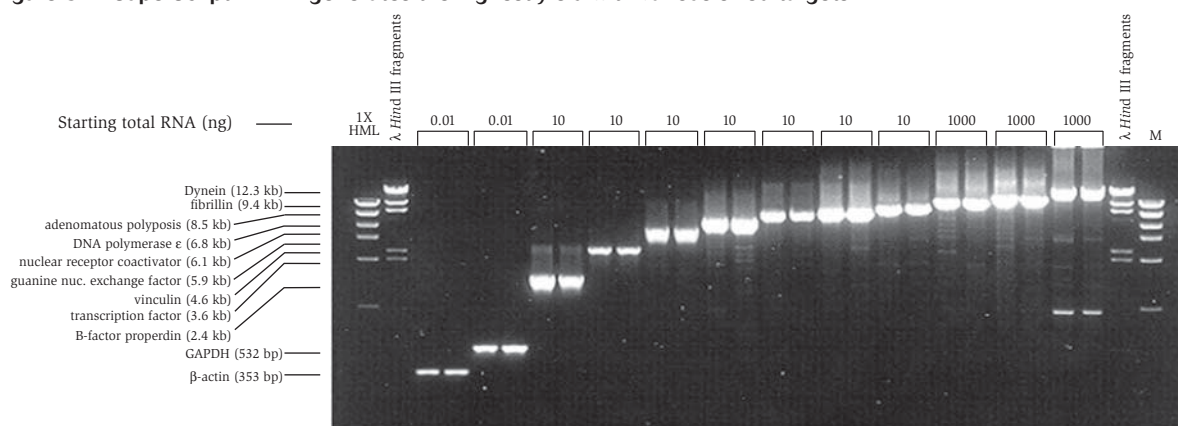
The SuperScript™ III First-Strand Synthesis System for RT-PCR delivers the outstanding performance of SuperScript™ III RT in an easy-to-use kit. You get all necessary components for first-strand cDNA synthesis, making it easy for novice and experienced users alike

**Figure 33 - SuperScript™ III compared to SuperScript™ II and M-MLV RTs**



An autoradiograph is shown of <sup>32</sup>P-labeled cDNA synthesized from a mixture of 0.25 µg of RNA each of 1.35 kb, 2.4 kb, 4.4 kb, 7.5 kb, and 9.5 kb with 200 units of each RT at various temperatures. Lane M is <sup>32</sup>P-labeled 1 kb DNA ladder.

**Figure 34 - SuperScript™ III RT generates the highest yield with various-sized targets**



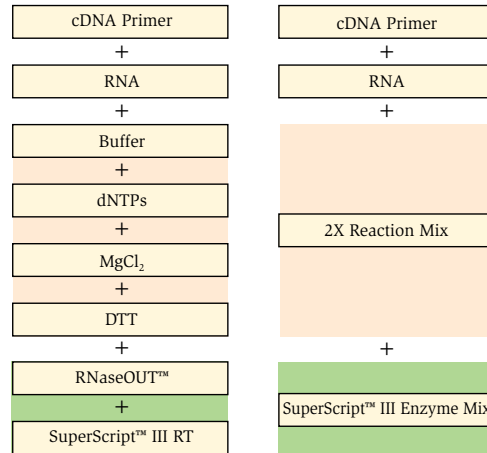
cDNA was synthesized from HeLa total RNA or rat total RNA for Dynein with oligo(dT) primer using 400 U of SuperScript™ III RT at 50°C. 10% of cDNA reaction was added to 50-µl PCR reaction containing primers for each gene and 2 U of Platinum® Taq DNA Polymerase or 1 U of Platinum® Taq DNA Polymerase High Fidelity, 35 or 40 cycles, 1 min/kb.

Product	Quantity	Cat. no.
SuperScript™ III Reverse Transcriptase (200 U/µl)	2000 U	18080-093
	10,000 U	18080-044
	4 x 10,000U	18080-085
SuperScript™ III First-Strand Synthesis System for RT-PCR	50 rxns	18080-051
Oligo(dT) <sub>20</sub> Primer	15 µg (50 rxns)	18418-020

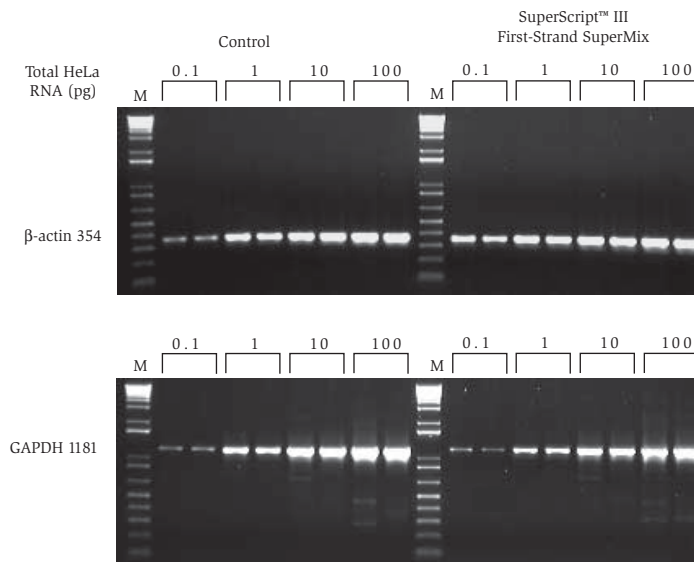
## High cDNA yields in a high-throughput format

The SuperScript™ III First-Strand Synthesis SuperMix provides high yields of first-strand cDNA in a convenient high-throughput supermix. The system is formatted with just two tubes—a 2X Reaction Mix and a SuperScript™ III Enzyme Mix—for simple, time-saving reaction set-up (Figure 35). An optimized annealing buffer is included for improved yields and sensitivity. The use of SuperScript™ III Reverse Transcriptase ensures higher cDNA yields, excellent sensitivity (Figure 36) and specificity, and compatibility with a wide variety of RT-PCR applications.

**Figure 35 - SuperScript™ III First-Strand Synthesis SuperMix provides convenient and time-saving reaction set-up**



**Figure 36 - SuperScript™ III First-Strand Synthesis SuperMix provides excellent sensitivity**



cDNA reactions were performed at 50°C using the SuperScript™ III First-Strand Synthesis SuperMix, 0.1-100 pg of total HeLa RNA, and primer sets for the housekeeping genes indicated. PCR reactions were performed at 1 min/kb for 40 cycles using Platinum® PCR SuperMix. The SuperScript™ III First-Strand Synthesis System for RT-PCR was used to synthesize cDNA for the control reactions.

Product	Quantity	Cat. no.
SuperScript™ III First-Strand Synthesis SuperMix	50 rxns	18080-400



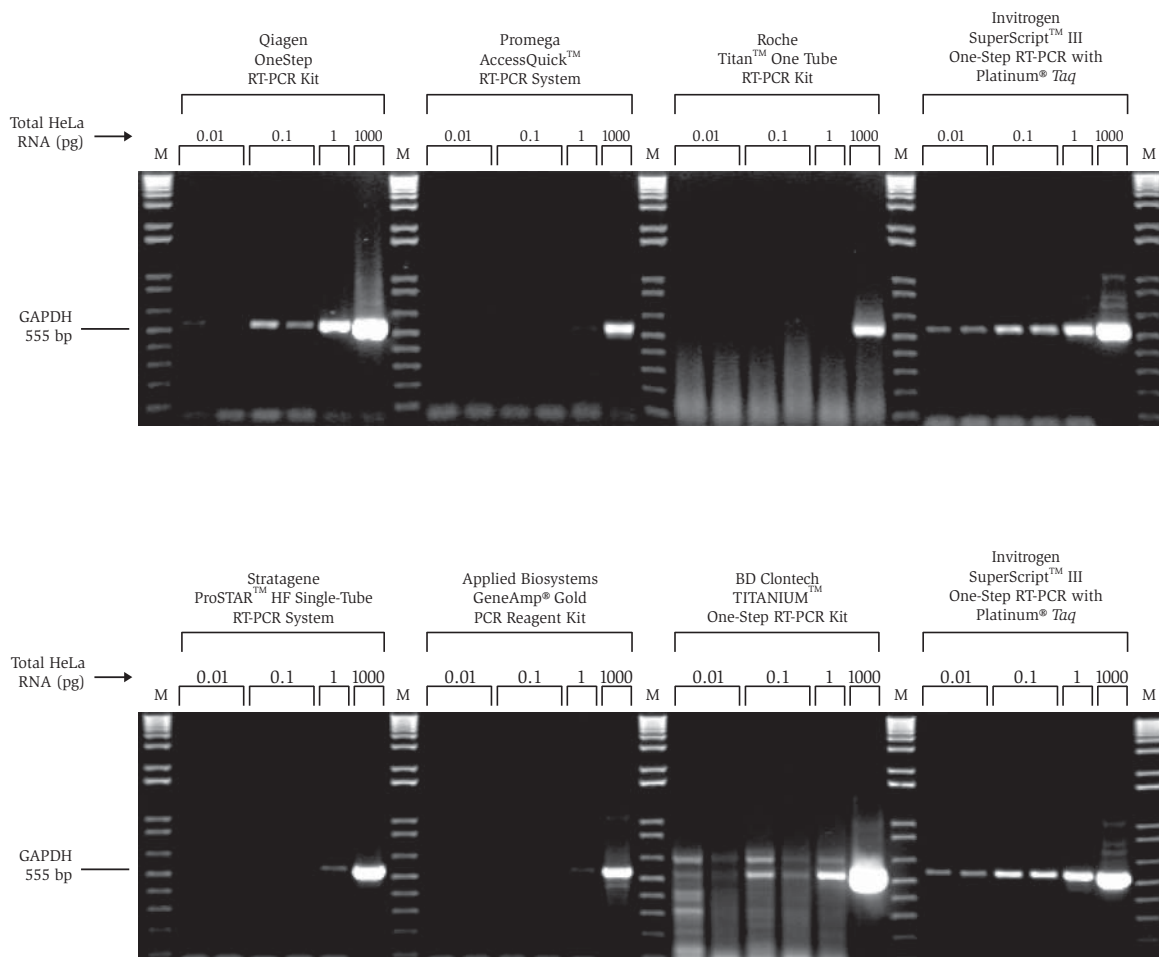
## The most sensitive one-step RT-PCR

The SuperScript™ III One-Step RT-PCR System with Platinum® *Taq* DNA Polymerase offers the highest sensitivity available for end-point detection. The system combines the thermostability of SuperScript™ III RT with the specificity of Platinum® *Taq* DNA Polymerase to provide greater priming specificity, higher product yields, and detection of a larger range of targets—all in an easy, one-step format.

With the SuperScript™ III One-Step RT-PCR System, you get:

- Routine detection down to 0.01 pg total RNA—the most sensitive one-step system available (Figure 37)
- cDNA synthesis up to 60°C with SuperScript™ III RT for greater success with RNA secondary structure
- Amplification of targets up to 4.5 kb in length for greater flexibility in your experiments

Figure 37 - The SuperScript™ III One-Step RT-PCR System with Platinum® *Taq* provides the highest sensitivity available



One-Step RT-PCR reactions were performed with 0.01, 0.1, 1.0 pg, and 1.0 ng total HeLa RNA using reagents and conditions specified in each manufacturer's protocol. The PCR annealing temperature was 55°C for all reactions.

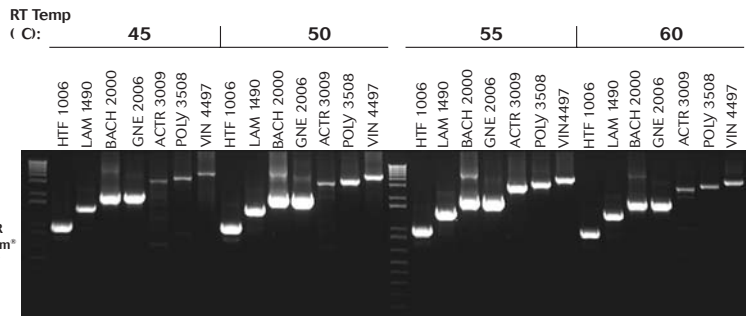
Product	Quantity	Cat. no.
SuperScript™ III One-Step RT-PCR System with Platinum® <i>Taq</i> DNA Polymerase	25 rxns	12574-018
	100 rxns	12574-026

## High-fidelity one-step RT-PCR for cloning

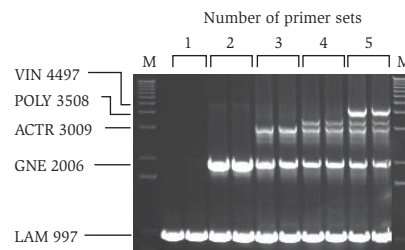
The SuperScript™ III One-Step RT-PCR System with Platinum® *Taq* High Fidelity provides high product yields and fidelity for applications such as cloning, gene detection, and quantitation. The enhanced thermostability of SuperScript™ III RT ensures superior cDNA yields, while the addition of Platinum® *Taq* DNA Polymerase High Fidelity delivers full-length, high-fidelity PCR for targets up to 9 kb in length. The SuperScript™ III One-Step RT-PCR Kit with Platinum® *Taq* High Fidelity offers:

- High RT-PCR product yields across a range of temperatures (Figure 38)
- Sensitive detection from as little as 1 pg total HeLa RNA
- A simple one-tube, one-step reaction format
- Compatibility with multiple applications including multiplex RT-PCR and cloning (Figure 39)
- Superior performance versus the competition (Figure 40)

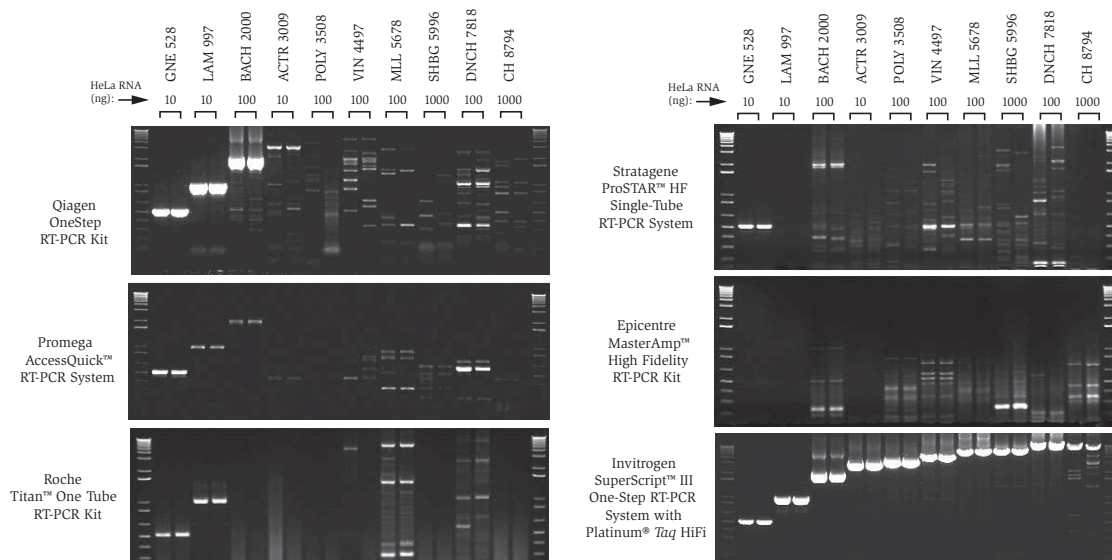
**Figure 38 - SuperScript™ III One-Step RT-PCR System with Platinum® *Taq* High Fidelity provides higher yields across a range of temperatures**



**Figure 39 - SuperScript™ III One-Step RT-PCR System with Platinum® *Taq* High Fidelity provides efficient multiplexing**



**Figure 40 - The SuperScript™ III One-Step RT-PCR System with Platinum® *Taq* High Fidelity outperforms the competition**



One-step RT-PCR reactions were performed according to each manufacturer's recommendations using the primers and starting total HeLa RNA quantities indicated.

Product	Quantity	Cat. no.
SuperScript™ III One-Step RT-PCR System with Platinum® <i>Taq</i> High Fidelity	25 rxns	12574-030
	100 rxns	12574-035

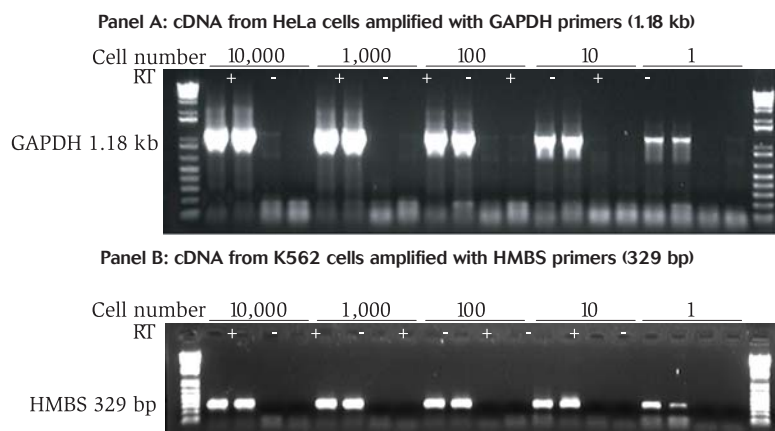
## High cDNA yields directly from cells

The SuperScript™ III CellsDirect™ cDNA Synthesis Kit provides high yields of first-strand cDNA from as little as one cell, minimizing sample loss from RNA isolation procedures and ensuring accurate representation of all transcripts. You can lyse mammalian cells, DNase treat, and synthesize cDNA in a single tube. The system uses SuperScript™ III RT to synthesize cDNA with increased specificity, produce higher yields of cDNA, and synthesize full-length products up to 4.5 kb.

The SuperScript™ III CellsDirect™ cDNA Synthesis Kit offers:

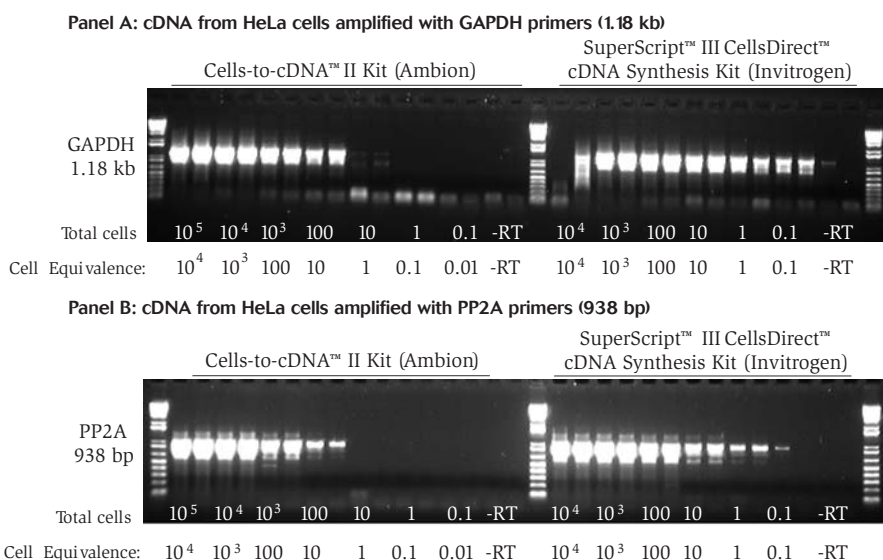
- High-yield cDNA synthesis from 1 to 10,000 cells (Figure 47)
- Time savings—go from cells to cDNA in less than two hours
- Simple format without RNA purification eliminates sample loss and enables detection of rare transcripts
- Compatibility with a wide variety of applications
- Superior performance versus the competition (Figure 48)

**Figure 47 - The SuperScript™ III CellsDirect™ cDNA Synthesis Kit provides linear and robust cDNA yields from 1 to 10,000 cells**



cDNA was synthesized directly from serial dilutions of cells (10,000 cells to 1 cell) using the SuperScript™ III CellsDirect™ cDNA Synthesis Kit. 2 µl of each cDNA synthesis was used in a 50-µl PCR reaction. 20% of the PCR reaction was analyzed by electrophoresis on a 1% agarose gel.

**Figure 48 - The SuperScript™ III CellsDirect™ cDNA Synthesis Kit outperforms the competition**



cDNA was synthesized from serial dilutions of HeLa cells using either the SuperScript™ III CellsDirect™ cDNA Synthesis Kit (Invitrogen) or the Cells-to-cDNA™ II Kit (Ambion) according to each manufacturer's specifications. 10% of each cDNA reaction was used in a 50-µl PCR reaction. 20% of the PCR reaction was analyzed by electrophoresis on a 1% agarose gel. Note: Due to protocol differences between manufacturers, cell numbers are provided to show both total cells and cell equivalents used in cDNA synthesis.

Product	Quantity	Cat. no.
SuperScript™ III CellsDirect™ cDNA Synthesis Kit	25 rxns	18080-200
	100 rxns	18080-300

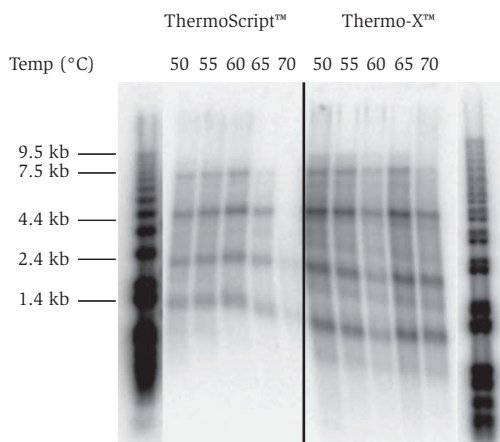
## The highest thermostability RT for GC-rich targets

Thermo-X™ Reverse Transcriptase provides the highest thermostability of any reverse transcriptase on the market to address your most challenging templates. With a half-life over 100 minutes at 65°C, Thermo-X™ RT is the best choice for sensitive and specific cDNA synthesis of difficult targets with high GC content or extensive secondary structure.

Thermo-X™ RT gives you:

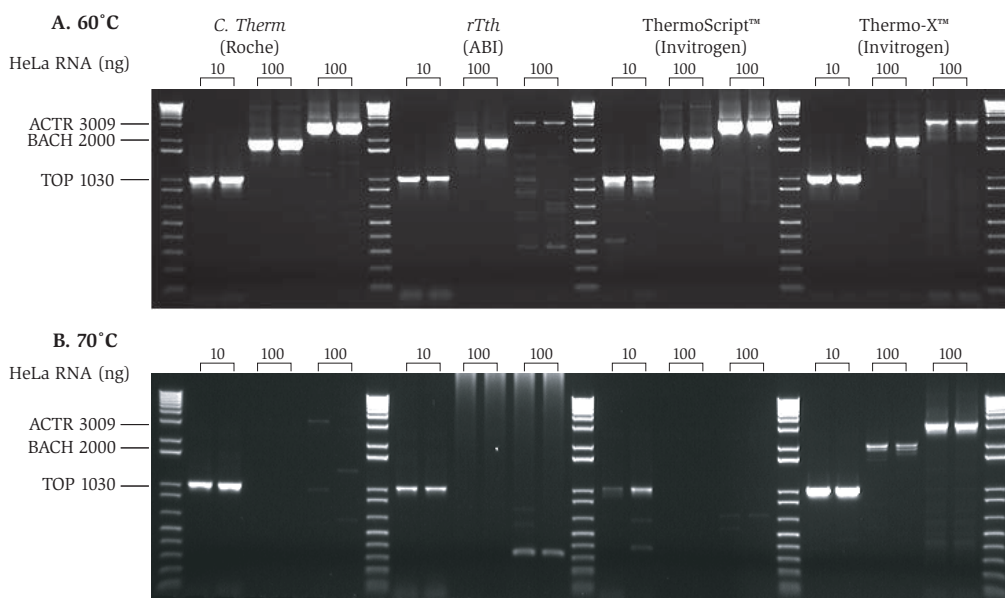
- High yields of cDNA at 65°C (Figure 49)
- Half-life of 120 minutes at 65°C for better results with high GC templates
- Sensitivity to detect as little as 1 pg total RNA
- Up to 12 kb target length capacity
- Superior performance versus the competition (Figure 50)

**Figure 49 - cDNA synthesis profile of ThermoScript™ RT and Thermo-X™ RT**



RT reactions were performed at 50°C-70°C, using 200 U of ThermoScript™ or Thermo-X™ RT in the presence of a poly(A)-tailed RNA ladder, oligo (dT)<sub>20</sub>, and α-<sup>32</sup>P dCTP. Reaction products were separated on a 1.2% alkaline agarose gel and visualized by autoradiography.

**Figure 50 - Thermo-X™ Reverse Transcriptase outperforms the competition at high temperatures**



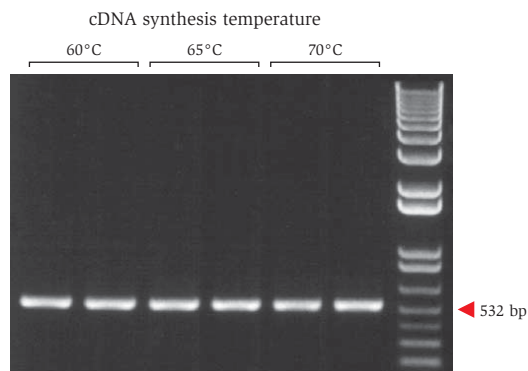
RT reactions were performed according to the manufacturers' instructions at 60°C or 70°C, using 10-100ng of total HeLa RNA and gene-specific primers. PCR reactions were performed for 35 cycles at 1 kb/min using Platinum® PCR SuperMix High Fidelity and indicated primer sets.

Product	Quantity	Cat. no.
Thermo-X™ Reverse Transcriptase	25 rxns	11150-025
	100 rxns	11150-100

## Successful RT-PCR of Difficult Small Targets

ThermoScript™ RT, an avian-derived enzyme with reduced RNase H activity, offers exceptional performance for difficult RNA templates. ThermoScript™ stays active at temperatures up to 70°C (Figure 51), so you get high specificity cDNA synthesis and amplification of problematic RNA templates with extensive secondary structure (Figure 52). To ensure best performance, try the ThermoScript™ RT-PCR System for First-Strand cDNA Synthesis. It's a complete kit, saving you time and effort in ordering and testing reagents.

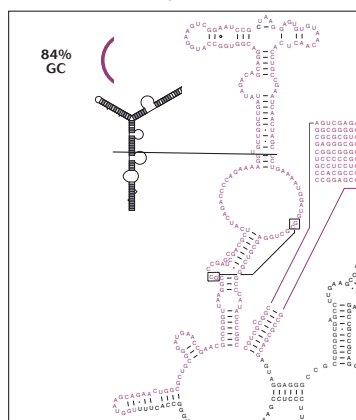
**Figure 51 - High yields at elevated temperatures**



cDNA was synthesized from total HeLa RNA with ThermoScript™ RT and oligo(dT) primers. 35 cycles of PCR were performed with Platinum® *Taq* DNA Polymerase and primers for phosphatase PP2A. Yield was maintained as the temperature was increased.

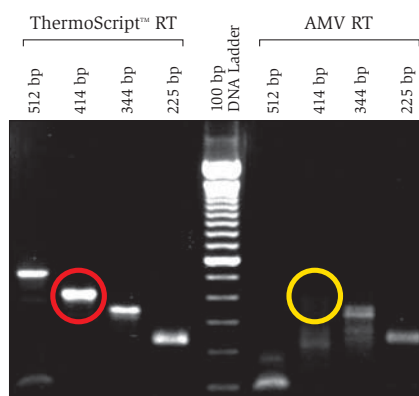
**Figure 52 - RT-PCR products from GC-rich ribosomal RNA with extensive secondary structure**

**A: Secondary structure**



Large subunit ribosomal RNA-5' half, 414 bp template region (in purple).

**B: RT-PCR results**



cDNA was synthesised from 10 ng total RNA using 15 units of ThermoScript™ RT at 55°C or 15 units AMV RT at 50°C. One tenth of the reaction was amplified with 1 unit of Platinum® *Taq* DNA Polymerase High Fidelity at 94°C for 3 min, then 40 cycles at 97°C for 30 s, 60°C for 30 s, and 68°C for 2-min. The 84% GC template was successfully amplified with ThermoScript™ RT.

Product	Quantity	Cat. no.
ThermoScript™ RT (15 U/μl)	25 rxns	12236-014
	100 rxns	12236-022
ThermoScript™ RT-PCR System for First-Strand cDNA Synthesis	25 rxns	11146-024
	100 rxns	11146-016
ThermoScript™ RT-PCR System and Platinum® <i>Taq</i> DNA Polymerase	25 rxns	11146-057
	100 rxns	11146-032
ThermoScript™ RT-PCR System and Platinum® <i>Taq</i> DNA Polymerase High Fidelity	100 rxns	11146-040
RT-PCR Primer and Control Set	20 rxns	10929-016

**Table 7 – Additional Reverse Transcriptase Products**

Product	Quantity	Cat. no.
SuperScript™ II Reverse Transcriptase	2,000 U	18064-022
	10,000 U	18064-014
	4 x 10,000 U	18064-071
SuperScript™ II One-Step RT-PCR System with Platinum® <i>Taq</i> DNA Polymerase	25 rxns	10928-034
	100 rxns	10928-042
SuperScript™ II One-Step RT-PCR System with Platinum® <i>Taq</i> DNA Polymerase High Fidelity	25 rxns	11922-010
	100 rxns	11922-028
SuperScript™ II First-Strand Synthesis System for RT-PCR	50 rxns	11904-018
MMLV Reverse Transcriptase	40,000 U	28025-013
	200,000 U	28025-021
Cloned AMV Reverse Transcriptase	750 U	12328-019
	3000 U	12328-027
Cloned AMV First-Strand cDNA Synthesis Kit	25 rxns	12328-032
	100 rxns	12328-040
RT-PCR Primer and Control Set	20 rxns	10929-016
RNaseOUT™ Recombinant Ribonuclease Inhibitor	5,000 U	10777-019
Random Primers	9 A260 units	48190-011
Oligo(dT) <sub>12-18</sub> Primer	25 µg	18418-012
Oligo(dT) <sub>20</sub> Primer	15 µg	18418-020
Taq DNA Polymerase PCR Buffer	2 x 1.25 ml	18067-017
MMLV Reverse Transcriptase Buffer	1 ml	18057-018
Ribonuclease H	30 U	18021-014
	4 x 30 U	18021-071
DNase I, amplification grade	100 U	18068-015

## Rely on the Invitrogen PCR and RT-PCR resource

---

These products may be covered by one or more Limited Use Label Licenses (See the Invitrogen catalog or our web site, [www.invitrogen.com](http://www.invitrogen.com)). By the use of these products you accept the terms and conditions of all applicable Limited Use Label Licenses. For research use only. Not intended for any animal or human therapeutic or diagnostic use. Printed in the U.S.A. ©2004 Invitrogen Corporation. All rights reserved. Reproduction forbidden without permission.

*PfuTurbo*<sup>®</sup> and *Herculase*<sup>®</sup> are registered trademarks of Stratagene. *PfuUltra*<sup>™</sup>, *Taq2000*<sup>™</sup>, *ProSTAR*<sup>™</sup>, and *AccuType*<sup>™</sup> are trademarks of Stratagene. *DNAzol*<sup>®</sup> and *TRIZol*<sup>®</sup> are registered trademarks of Molecular Research Center, Inc. *AccessQuick*<sup>™</sup> is a trademark of Promega Corporation. *AmpliQ*<sup>®</sup> and *GeneAmp*<sup>®</sup> Gold are registered trademarks of Applied Biosystems. *HotStarTaq*<sup>®</sup> is a registered trademark of the Qiagen Group. *JumpStart*<sup>™</sup> and *AccuTaq*<sup>™</sup> are trademarks of Sigma. *Advantage*<sup>™</sup> and *TITANIUM*<sup>™</sup> are trademarks of BD Biosciences. *WAVE* and *DNASep* are trademarks or registered trademarks of Transgenomic, Inc. *Expand*<sup>™</sup>, *Titan One Tube*<sup>™</sup> and *FastStart*<sup>™</sup> are trademarks of a member of the Roche Group. *MasterAmp*<sup>™</sup> is a trademark of Epicentre. *Ready-To-Go*<sup>™</sup> is a trademark of Amersham Biosciences. *Cells-to-cDNA*<sup>™</sup> is a trademark of Ambion<sup>®</sup>. *Ex Taq*<sup>™</sup> is a trademark of Takara Mirus Bio.

*AccuPrime*<sup>™</sup>, *GeneRacer*<sup>™</sup>, *Lipofectamine*<sup>™</sup>, *LUX*<sup>™</sup>, *Micro-to-Midi*<sup>™</sup>, *SuperScript*<sup>™</sup>, *ThermalAce*<sup>™</sup>, *ThermoScript*<sup>™</sup>, *Thermo-X*<sup>™</sup>, *CellsDirect*<sup>™</sup> and *XCell SureLock*<sup>™</sup> are trademarks of Invitrogen Corporation. *E-Gels*<sup>®</sup>, *Elongase*<sup>®</sup>, *Gateway*<sup>®</sup>, *Novex*<sup>®</sup>, *Platinum*<sup>®</sup>, *TA Cloning*<sup>®</sup>, and *TOPO*<sup>®</sup> are registered trademarks of Invitrogen Corporation. *SYBR*<sup>®</sup> is a registered trademark of Molecular Probes.





**Corporate headquarters:**

1600 Faraday Avenue • Carlsbad, CA 92008 USA • Tel: 760 603 7200 • Fax: 760 602 6500 • Toll Free Tel: 800 955 6288 • E-mail: [tech\\_service@invitrogen.com](mailto:tech_service@invitrogen.com) • [www.invitrogen.com](http://www.invitrogen.com)

**European headquarters:**

Invitrogen Ltd • Inchinnan Business Park • 3 Fountain Drive • Paisley PA4 9RF, UK • Tel: +44 (0) 141 814 6100 • Fax: +44 (0) 141 814 6260 • E-mail: [eurotech@invitrogen.com](mailto:eurotech@invitrogen.com)