User Manual

Advantage®-HF 2 PCR Kit User Manual

Clontech

United States/Canada
800.662.2566
Asia Pacific
+1.650.919.7300
Europe
+33.(0)1.3904.6880
Japan
+81.(0)77.543.6116

Clontech Laboratories, Inc.
A Takara Bio Company
1290 Terra Bella Ave.
Mountain View, CA 94043
Technical Support (US)
E-mail: tech@clontech.com
www.clontech.com

PT3317-1 (PR15722)
Published: 14 May 2001

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I. Introduction

The **Advantage®-HF 2** (High-Fidelity) **PCR Kit** is a TITANIUM *Taq*-based system designed to amplify cDNA or genomic templates with exceptionally high fidelity.

Why use Advantage®-HF 2?

While proofreading polymerases such as *Pfu* can offer high -fidelity amplification, they suffer from low efficiencies in many PCR applications; as a result, they may not produce sufficient quantities of full-length product. In contrast, the Advantage-HF 2 PCR Kit, which can be used to amplify fragments up to about 3.5 kb, offers both high fidelity and high efficiency.

The Advantage-HF 2 Polymerase Mix is a high-performance PCR system that combines TITANIUM *Taq* DNA Polymerase—a nuclease-deficient N-terminal deletion of *Taq* DNA polymerase plus TaqStart Antibody to provide automatic hot-start PCR (Kellogg *et al.*, 1994)—and a minor amount of a proofreading polymerase. The dual-polymerase system provides both high sensitivity and flexibility in amplifying a wide range of DNA templates, and TaqStart provides automatic hot-start PCR.

The Advantage-HF 2 Kit's high-fidelity feature was developed as described by Ling *et al.* (1991) and Eckert & Kunkel (1990, 1991). We have improved fidelity further by making additional modifications to several components in the Kit (

The high-fidelity advantage

We used a fidelity assay to measure the accuracy of the HF 2 Kit in comparison to *Taq* polymerase and our Advantage 2 Kit. This assay measures nucleotide misincorporation and is based on amplification of a gene encoding an *E. coli* ribosomal protein (Mo *et al.*, 1991). Mutations in this gene often confer streptomycin resistance on the host. Upon introduction of the amplified DNA into *E. coli*,

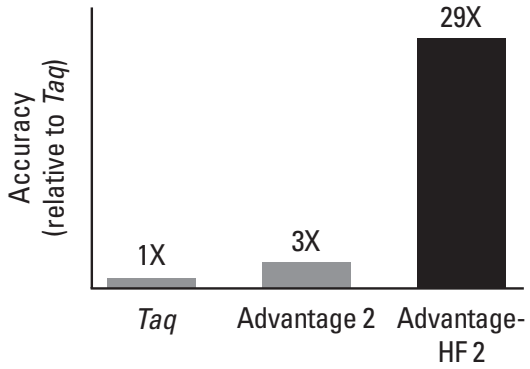

Figure 1. Advantage®-HF 2 is 29 times more accurate than *Taq*. Assay is based on 25 cycles of amplification (see text). The error rate of *Taq* polymerase was 2.75×10^{-5} $bp^{-1}cycle^{-1}$, determined by DNA sequencing.

I. Introduction *continued*

TABLE I. FIDELITY OF ADVANTAGE®-HF 2 BASED ON SEQUENCING DATA

Enzyme	Error rate [a] (per 100,000 bp)
Taq	66 [b]
Advantage 2	24
Advantage-HF 2	2.3

[a] determined with individual clones after 25 PCR cycles.
[b] agrees with published data (Ling *et al.*, 1991; Cariello *et al.*, 1991)

the ratio of total transformants to streptomycin-resistant transformants provides a comparative measure of PCR fidelity (Figure 1). The fidelity of Advantage-HF 2 was normalized by sequencing (Table I). The high level of fidelity delivered by the Advantage-HF 2 system increases confidence in sequence derived from PCR products and is beneficial in a variety of PCR applications, including expression studies of amplified full-length cDNAs, generation of cDNA libraries, and analysis of homologous genes amplified with degenerate primers.

High-fidelity amplification of cDNA and genomic templates

Advantage-HF 2 was used to amplify several cDNA and genomic templates of different lengths (Figure 2). Note that Lane 5 shows amplification of a relatively long template (about 3.5 kb) amplified from calf thymus DNA.

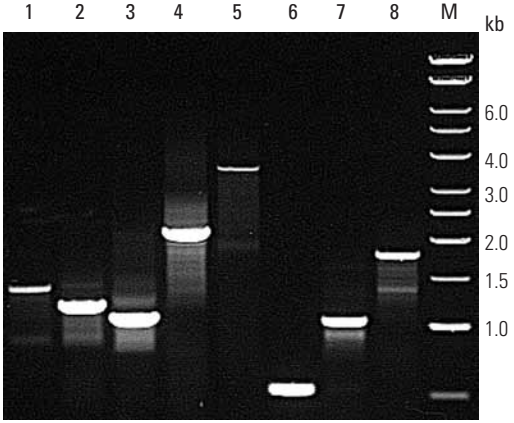

Figure 2. Amplification of a wide size range of cDNA and genomic fragments using Advantage®-HF 2. Lanes 1–2: amplification from Human Placenta Marathon-Ready cDNA. Lane 1: 1.3-kbTFR fragment. Lane 2: 1.2-kb β-actin fragment. Lanes 3–5: amplification of several size fragments (1.0–3.5 kb) of the BPTI gene from calf thymus DNA. Lanes 6–8: amplification of several fragments (0.5–1.8 kb) of the human cardiac β-myosin heavy chain from human genomic DNA. Lane M: 1-kb DNA size markers.

I. Introduction *continued*

Increase elongation efficiency for longer fragments

Two reaction buffers are included in the Advantage-HF 2 Kit: the HF 2 Buffer and the original Advantage 2 PCR Buffer. Using the HF 2 Buffer will deliver the highest possible fidelity for amplifying fragments up to ~3.5 kb. To amplify longer fragments, you can improve elongation efficiency by combining the HF 2 and Advantage 2 buffers in varying proportions. However, some fidelity will be sacrificed.

Automatic hot start with TaqStart® Antibody

TaqStart is a neutralizing monoclonal antibody that recognizes both native and N-terminal deletion mutant forms of *Taq* polymerase. It is included in the Advantage-HF 2 Polymerase Mix for automatic hot-start PCR. In antibody-mediated hot start, the antibody binds and inhibits the polymerase at ambient temperatures during PCR reaction setup. When the PCR reaction mix reaches the high temperatures required for the onset of thermal cycling, however, the antibody is denatured, restoring polymerase activity and allowing amplification to commence. Restoration is complete and irreversible, so the polymerase retains full enzymatic activity throughout the process of amplification.

TaqStart significantly improves the efficiency and specificity of PCR amplifications by reducing background DNA synthesis at ambient temperatures, before thermal cycling begins (Kellogg *et al*., 1994; *Clontechniques,* April 1994). Specifically, this antibody reduces or eliminates nonspecific amplification products and primer-dimer artifacts created prior to the onset of thermal cycling due to nonspecific polymerase activity.

Besides increased specificity and sensitivity, the built-in hot start in the Advantage-HF 2 Polymerase Mix offers convenience. Other methods of hot start require extra steps such as adding and premelting wax beads, or adding a critical component after the initial denaturation. These extra steps are inconvenient and introduce a potential source of cross-contamination. In contrast, because TaqStart is already included in the polymerase mix, it provides hot-start PCR with virtually no risk of cross-contamination.

Which Advantage® Kit should I use?

Advantage-HF 2 can be used to amplify moderately sized templates when exceptionally high fidelity is required. It is the recommended kit if your PCR product will be used for applications such as cloning or mutation analysis. For applications that do not require such high fidelity, use one of our other Advantage Kits described below.

For amplifying **GC-rich templates:** The Advantage-GC 2 PCR Kit (Cat. Nos. 639119, 639120) and Polymerase Mix (Cat. No. 639114) efficiently amplify moderate-complexity sequences with high GC content, such as the 5' ends of many mammalian cDNAs, that cannot be amplified using conventional PCR techniques.

I. Introduction *continued*

For amplifying **GC-rich genomic** templates, use our Advantage-GC Genomic Kit (Cat. Nos. 639117, 639118). These kits are recommended for use with Clontech's GenomeWalker Kits (Cat. Nos. 638901, 638902, 638903, 638904, and several LD-Insert Screening Amplimer Sets.

For amplifying **non–GC-rich genomic** templates, use the Advantage Genomic PCR Kit (Cat. Nos. 639103, 639104) or Polymerase Mix (Cat. No. 639105).

For **long-distance PCR** of moderate-complexity templates, or for special PCR applications such as RACE, cDNA subtraction, or differential display, use the Advantage 2 PCR Kit (Cat. Nos. 639207, 639206) or Polymerase Mix (Cat. Nos. 639201, 639202).

For any other general-purpose PCR, use our TITANIUM *Taq* Polymerase Mixes (Cat. Nos. 639208, 639209) and Kits (Cat. Nos. 639210, 639211).

I. List of Components

Advantage®-HF 2 PCR Kit (Cat. Nos. 639124 [10 rxns], 639123 [100 rxns])

Store all reagents at –20°C.

10 rxns **100 rxns**

- **10 µl** **100 µl** **50X Advantage-HF 2 Polymerase Mix**

  Includes TITANIUM *Taq* DNA Polymerase—a nuclease-deficient N-terminal deletion of *Taq* DNA polymerase plus TaqStart Antibody (1.1 µg/µl)—and a minor amount of a proofreading polymerase in the following storage buffer.

Concentration in 50X mix	Component	Final rxn. concentration
50 %	Glycerol	1.0 %
15 mM	Tris-HCl (pH 7.5)	0.3 mM
75 mM	KCl	1.5 mM
1 mM	EDTA	20 µM

- **60 µl** **600 µl** **10X HF 2 PCR Buffer**
- **60 µl** **600 µl** **10X Advantage® 2 PCR Buffer**
- **60 µl** **600 µl** **10X HF 2 dNTP Mix**
- **400 µl** **4 ml** **PCR-Grade H_2O**
- **10 µl** **100 µl** **Control DNA Template**
  λ DNA (0.2 ng/µl)
- **10 µl** **40 µl** **Control Primer Mix** (10 µM each)
  The sequences are:

5' primer: 5'–TTGGTTGATCGTGGTGCAGAGAACGTTG–3'
3' primer: 5'–GAGAAGGTCACGAATGAACCAGGCGATAA–3'

III. Additional Materials Required

The following reagents are not supplied.

- [optional]: **Mineral oil** (We recommend Sigma Cat. No. M-3516)
- **0.5-ml PCR reaction tubes** (We recommend Perkin-Elmer GeneAmp 0.5-ml reaction tubes [Cat. No. N801-0737 or N801-0180].)
- **Thermal cycler** (Perkin-Elmer GeneAmp 9600 or equivalent)
- **Pipettors dedicated for PCR**
- **PCR pipette tips** suitable for use with the above pipettors and preferably equipped with hydrophobic filters.
- **DNA size markers** (See Section IV.E)
- **5X Stop/loading buffer** (Sambrook *et al.* [1989] provides several recipes.)

IV. Advantage®-HF 2 PCR Protocol

PLEASE READ ENTIRE PROTOCOL BEFORE STARTING.

A. General Considerations

1. Primer design

   Primer design is the single largest variable in PCR applications and the single most important factor in determining the success or failure of PCR reactions. *Always check and recheck your primer design before constructing or ordering primers.* Visit **alces.med.umn.edu/VGC.html** on the web for helpful guidelines on primer design.

   Primer length and GC content: In general, for amplifying GC-rich sequences, primers should have a T_m of at least 70°C to achieve optimal results in a two-step cycling program with a 68°C annealing/extension step. Therefore, whenever possible, primers should be *at least* 22 (nt) long (25–30-mers are preferred) and should have a GC content of 45–60%.

2. Template quality

   Because PCR amplification proceeds exponentially, many conventional PCR applications work well with templates of average or even low quality. In many applications, the Advantage-HF 2 Polymerase Mix will also tolerate a wide range of template quality.

   However, the longer or more complex the target, the more important template quality becomes. As target length increases, the number of unnicked, full-length targets decreases; poor-quality DNA has very few long targets that are unnicked. Furthermore, some depurination

IV. Advantage®-HF 2 PCR Protocol *continued*

occurs when DNA is denatured during thermal cycling, which can lead to truncated products. Therefore, it is particularly important that you prepare high-quality, high molecular-weight DNA when amplifying long targets.

3. Good PCR practices
   a. Prepare reactions with dedicated pipettors in a dedicated workspace

      Due to the tremendous amplification power of PCR, minute amounts of contaminating DNA can produce nonspecific amplification; in some instances, contaminants can cause DNA bands even in the absence of added template DNA. We recommend that you use small aliquots of starting material to avoid contaminating your stocks. You should set up your PCR reactions in a dedicated lab area or noncirculating containment hood and use dedicated pipettors, PCR pipette tips with hydrophobic filters, and dedicated solutions. Finally, perform post-PCR analysis in a separate area using a separate set of pipettors.

   b. Pipetting

      Because of the small volumes used in PCR experiments and the potential for tube-to-tube variation, careful pipetting technique is extremely important. Always be sure that no extra solution is on the outside of the pipette tip before transfer. When adding solution to a tube, immerse the tip into the reaction mixture, deliver the contents from the pipette tip into the mixture, and pipet up and down several times.

   c. Use a Master Mix

      A Master Mix contains the appropriate volumes of all reagents required for multiple PCR reactions. It saves time and greatly reduces tube-to-tube variation. If multiple templates are being tested with the same primers, include the primers in the Master Mix. If one template is being tested with multiple primer sets, include the template in the Master Mix. If you are setting up several sets of parallel samples, assemble multiple Master Mixes (e.g., each with a different set of primers). *The Master Mix should be thoroughly mixed before use (i.e., vortexed without bubbling).*

   d. Always include positive and negative controls (i.e., H_2O instead of DNA template).

4. Touchdown PCR

   "Touchdown" PCR can significantly improve the specificity of many PCR reactions in a wide variety of applications (Don *et al.*, 1991; Roux, 1995). Briefly, touchdown PCR involves using an annealing/extension temperature that is several degrees (typically 3–10°C) higher than the T_m of the primers during the initial PCR cycles (typically 5–10 cycles).

IV. Advantage®-HF 2 PCR Protocol *continued*

The annealing/extension temperature is then reduced to the primer T_m for the remaining PCR cycles.

5. TaqStart Antibody provides automatic hot-start PCR

   The use of a manual hot start or wax bead-based hot start is not required when using Advantage -HF 2. As discussed in the Introduction, hot start is automatic because the enzyme mix already contains TaqStart Antibody.

6. Use of additives

   TaqStart Antibody binds N-terminal deletion mutant *Taq* polymerase with high affinity under the conditions described in the protocol. Adding DMSO to the reaction buffer will not interfere with TaqStart function. However, the addition of formamide or other cosolvents may disrupt TaqStart function. Furthermore, excessive glycerol, solutes (e.g., salts), pH extremes, or other deviations from the recommended reaction conditions may reduce the effectiveness of the antibody and/or DNA polymerases.

7. Advantage-HF 2 is not intended for certain applications

   Because of the improved fidelity from long and accurate PCR, Advantage-HF 2 is not recommended for mutagenesis protocols involving so-called "sloppy" PCR.

IV. Advantage®-HF 2 PCR Protocol *continued*

B. Control PCR Reactions

The following PCR reactions can be performed in parallel with your experiments as controls to ensure that the Advantage-HF 2 Kit is working properly. A positive control template and primers are provided in the kit.

1. Place all components on ice and allow to thaw completely. Mix each component thoroughly before use.
2. Combine the following reagents in a 0.5-ml PCR tube.

Positive Control	Negative Control	
32 µl	37 µl	PCR-Grade H_2O
5 µl	5 µl	10X HF 2 PCR Buffer
5 µl	---	Control DNA Template (~0.2 ng/µl)
2 µl	2 µl	Control Primer Mix (10 µM ea.)
5 µl	5 µl	10X HF 2 dNTP Mix
1 µl	1 µl	50X Advantage-HF 2 Polymerase Mix
50 µl	**50 µl**	**Total volume**

3. Mix well and spin briefly to collect all the liquid in the bottom of the tube.
4. If your thermal cycler does not have a "hot lid", overlay 1–2 drops of mineral oil onto the reaction mixture to prevent evaporation during cycling. A good seal of mineral oil should have a well-defined meniscus between the two phases. Cap the PCR tubes firmly.
5. Commence thermal cycling. Use the cycling parameters described in Section C below. 20–22 cycles with a 4-min annealing/extension time is sufficient for amplification of the positive control template provided in the kit.
6. Transfer a 5-µl sample of your PCR reaction to a fresh tube and add 1 µl of 5X stop/loading buffer. Analyze your sample(s), along with suitable DNA size markers, by electrophoresis on a 0.8–1.2 % agarose/EtBr gel.

Expected results: If you are using the positive control reagents provided in the kit, the reaction should produce a single major band of 2 kb. No bands should be generated in the negative control.

C. Recommended Cycling Parameters

Use the following guidelines when setting up your initial experiments with the Advantage-HF 2 Polymerase Mix. These are general guidelines— optimal parameters may vary with different thermal cyclers and will depend on your particular primers, templates, and other experimental variables.

Note: When using the Advantage-HF 2 Kit with other kits, use the parameters recommended in the protocol for that kit.

IV. Advantage®-HF 2 PCR Protocol *continued*

Cycle Parameters

(PE 480)	(PE 9600)
• 94°C for 1 min	• 94°C for 15 sec
• 25–35 cycles [A] 94°C 30 sec [B] 68°C 4 min [C]	• 25–35 cycles [A] 94°C 5–15 sec [B] 68°C 4 min [C]
• 68°C for 3 min [D]	• 68°C for 3 min [D]

[A] 25 cycles for multiple-copy genes or medium-to-high abundance cDNAs; 30–35 cycles for single- or low-copy-number genes or rare cDNAs. For most applications, we prefer two-step cycles (denaturation at T_1 followed by annealing and extension at T_2) instead of three-step cycles (denaturation at T_1 followed by annealing at T_2 followed by extension at T_3). Three-step cycles will be necessary when the T_m of the primers is less than 60–65°C, and in certain special protocols.

[B] Use the shortest possible denaturation time. Exposure of DNA to high temperatures causes some depurination of single-stranded DNA during denaturation, which eventually leads to truncation. High temperature also leads to gradual loss of enzyme activity.

[C] Use the highest possible annealing/extension temperature. See Note A. Shorter targets may be amplified using shorter extension times.

We recommend using 1 min per kb of expected target.

[D] Optional: This final extension may reduce background in some cases.

D. Amplification of Longer Fragments with the Advantage® 2 Buffer

The Advantage-HF 2 Kit contains two buffers—the HF 2 Buffer and the standard Advantage 2 Buffer. When used with the HF 2 Buffer, this kit delivers the highest possible fidelity. Fragments of up to ~3.5 kb can be amplified under these conditions. To amplify longer fragments, some of the increase in fidelity can be sacrificed to improve elongation efficiency by combining the HF 2 and Advantage 2 buffers in varying proportions. To amplify longer fragments, we recommend replacing the smallest amount of HF 2 Buffer with Advantage 2 Buffer that allows satisfactory amplification. For example, a 6-kb fragment can be amplified from a cDNA library in a 50-µl PCR reaction containing 4 µl of 10X HF 2 Buffer (80% final) and 1 µl of 10X Advantage 2 Buffer, conditions allowing a ~3-fold increase in fidelity over the cDNA buffer (Figure 3). Optimal conditions for the amplification of other fragments should be determined individually. We recommend initially trying HF 2 Buffer concentrations in the 50–100% range for fragments up to 10 kb.

E. Recommendations for Electrophoresis

We recommend that you transfer a 5-µl sample of your PCR reaction to a fresh tube and add 1 µl of 5X stop/loading buffer. The remaining

IV. Advantage®-HF 2 PCR Protocol *continued*

45 µl of the reaction mixture can be subjected to further cycling if you do not see a product. Analyze your sample(s), along with suitable DNA size markers, by electrophoresis on a suitable agarose gel containing 0.1–0.5 µg/ml EtBr. The percentage agarose and the DNA size markers you choose will depend on the expected range of insert sizes. You may wish to refer to the following general guidelines before assembling your gel.

Recommendations for agarose gels:

Expected insert size range	Recommended % agarose	Recommended DNA size markers
0.3–1.5 kb	1.5	ϕX174/*Hae* III
0.5–10 kb	1.2	1-kb DNA ladder
>5 kb	0.8	λ/*Hin*d III

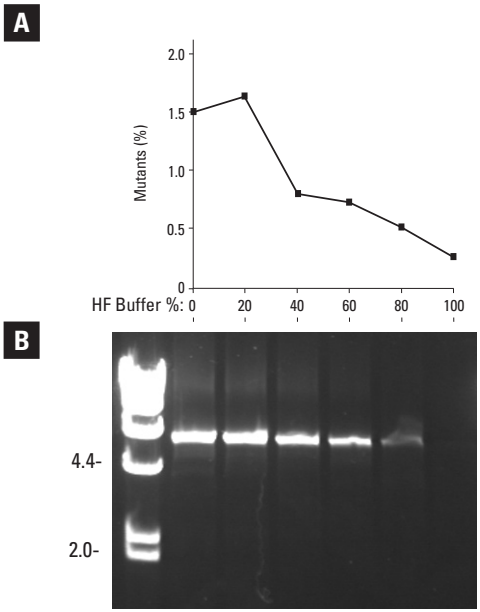

Figure 3. The effect of HF Buffer concentration on PCR fidelity (Panel A) and amplification (Panel B) of a 6.0-kb cDNA fragment from Human Placenta cDNA. See the Introduction for a description of the fidelity assay. Size markers are l/*Hin*d III DNA.

V. Troubleshooting Guide

The following **general** guidelines apply to most PCR reactions. However, no attempt has been made to address troubleshooting for all of the many applications for which the Advantage-HF 2 Kit can be used. When using the kit with another Clontech product, additional, application-specific troubleshooting information can be found in the relevant User Manual.

A. No product observed

PCR component missing or degraded	Use a checklist when assembling reactions. Always perform a positive control to ensure that each component is functional. If the positive control does not work, repeat the positive control only. If the positive control still does not work, repeat again replacing individual components to identify the faulty reagent.
Too few cycles	Increase the number of cycles (3–5 additional cycles at a time).
Annealing temp. too high	Decrease the annealing temperature in increments of 2–4°C.
Suboptimal primer design	Redesign your primer(s) after confirming the accuracy of the sequence information. If the original primer(s) was less than 22 nt long, try using a longer primer. If the original primer(s) had a GC content of less than 45%, try to design a primer with a GC content of 45–60%.
Not enough template	After trying more cycles, repeat PCR using a higher concentration of DNA.
Poor template quality	Check template integrity by electrophoresis on a standard TBE-agarose gel. If necessary, repurify your template using methods that minimize shearing and nicking.
Denaturation temp. too high or low	Optimize denaturation temperature by decreasing or increasing it in 1°C increments. A denaturation temperature that is too high can lead to degradation of the template, especially for long target sequences.
Denaturation time too long or too short	Optimize denaturation time by decreasing or increasing it in 10-sec increments. A denaturation time that is too long can lead to degradation of the template, especially for long target sequences.
Extension time too short	Especially with longer templates, increase the extension time in 1-min increments.
Too little enzyme	The Advantage-HF 2 Polymerase Mix is 50X for most applications. Therefore, try to optimize the cycle parameters as described above before increasing the

V. Troubleshooting Guide *continued*

	enzyme concentration. In rare cases, the yields can be improved by increasing the concentration of the enzyme mix. However, increasing the concentration >2X is likely to lead to higher background levels.
[Mg^{2+}] is too low	The Advantage-HF 2 Polymerase Mix performs well over a broad range of Mg^{2+} concentrations. Therefore, as long as you use the buffer included with the mix, it is unlikely that a lack of product is due to problems with the Mg^{2+} concentration. However, high concentrations of EDTA or other metal chelators in the template stock solution can reduce the effective concentration of Mg^{2+} to below a minimum level.
[dNTPs] not optimal	The Advantage-HF 2 PCR Kit contains a carefully balanced mixture of the four dNTPs. Replacement of this mixture with a standard dNTP mix (200 μM each) may improve the DNA yield, but may also result in lower fidelity.
Difficult target	Some targets are inherently difficult to amplify. In most cases, this is due to unusually high GC content and/or secondary structure. The Advantage-GC 2 Polymerase Mix and Kits are recommended in these situations.

B. Multiple products

Too many cycles	Reducing the cycle number may eliminate non-specific bands.
Annealing temp. too low	Increase the annealing/extension temperature in increments of 2–3°C.
Suboptimal primer design	Redesign your primer(s) after confirming the accuracy of the sequence information. If the original primer(s) was less than 22 nt, try using a longer primer. If the original primer(s) had a GC content of less than 45%, try to design a primer with a GC content of 45–60%.
Touchdown PCR needed	"Touchdown" PCR significantly improves the specificity of many PCR reactions in various applications (Don *et al.*, 1991; Roux, 1995). See Section IV.A.4 for more information.
Contamination	See Section D.

V. Troubleshooting Guide *continued*

C. Products are smeared

Too many cycles	Reduce the cycle number by 3–5 to see if non-specific bands disappear.
Denaturation temp. too low	Try increasing the denaturation temperature in increments of 1°C.
Extension time too long	Decrease the extension time in 1–2-min increments.
Poor template quality	Check template integrity by electrophoresis on a denaturing agarose gel. Repurify your template if necessary.
Touchdown PCR needed	See "Touchdown PCR needed" under previous section.
Too much enzyme	Advantage-HF 2 Polymerase Mix is 50X for most applications; however, a 1X final concentration of the enzyme mix may be too high for some applications. If smearing is observed, first try optimizing the cycle parameters as described above, then try reducing the enzyme concentration to 0.5–0.2X.
[Mg^{2+}] is too high	TITANIUM *Taq* has a broader Mg^{2+} optimum than native *Taq* DNA polymerase (i.e., it performs well over a wider range of [Mg^{2+}] with no loss of efficiency.) Therefore, as long as you have used the buffers supplied in the kit, it is unlikely that smearing is due to problems with the Mg^{2+} concentration. Altering the concentration of Mg^{2+} can result in lower fidelity.
Too much template	Try a lower concentration of DNA template in the PCR reaction.
Contamination	See Section D below.

D. Dealing with contamination

Contamination most often results in extra bands or smearing. Include a negative control (i.e., a control using deionized H_2O instead of the DNA template) in every PCR experiment to determine if the PCR reagents, pipettors or PCR reaction tubes are contaminated with previously amplified targets.

If possible, set up the PCR reaction and perform the post-PCR analysis in separate laboratory areas with separate sets of pipettors.

Laboratory benches and pipettor shafts can be decontaminated by depurination. Wipe surfaces with 1N HCl followed by 1N NaOH. Then neutralize with a neutral buffer (e.g., Tris or PBS) and rinse with H_2O.

V. Troubleshooting Guide *continued*

We advise using commercially available aerosol-free pipette tips.

There is an enzymatic method for destroying PCR product carryover (Longo *et al.*, 1990). It involves incorporation of dUTP into the PCR products and subsequent hydrolysis with uracil-N-glycosylase (UNG).

When performing PCR directly on phage plaques or bacterial colonies, failure to isolate single plaques or colonies will also produce multiple bands.

VI. References

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VII. Related Products

Product	Cat. No.
• Human GenomeWalker™ Kit	638901
• Mouse GenomeWalker™ Kit	638902
• Rat GenomeWalker™ Kit	638903
• Universal GenomeWalker™ Kit	638904
• Marathon-Ready cDNAs	many
• RT-PCR Amplimer Sets	many
• Advantage® RT-for-PCR Kit	639505 639506
• QUICK-Clone™ cDNAs	many
• TITANIUM™ *Taq* DNA Polymerase	639208 639209
TITANIUM™ *Taq* PCR Kit	639210 639211
• Advantage® 2 Polymerase Mix	639201 639202
Advantage® 2 PCR Kit	639206 639207
• Advantage® Genomic Polymerase Mix	639110
Advantage® Genomic PCR Kit	639103 639104
• Advantage®-GC 2 Polymerase Mix	639114
Advantage®-GC 2 PCR Kit	639119 639120
• TaqStart® Antibody	639250 639251