Taq Extender™ PCR Additive

INSTRUCTION MANUAL

Catalog #600148 (1000 U)
Revision #046001b

For In Vitro Use Only

LIMITED PRODUCT WARRANTY

This warranty limits our liability to replacement of this product. No other warranties of any kind, express or implied, including without limitation, implied warranties of merchantability or fitness for a particular purpose, are provided by Stratagene. Stratagene shall have no liability for any direct, indirect, consequential, or incidental damages arising out of the use, the results of use, or the inability to use this product.

ORDERING INFORMATION AND TECHNICAL SERVICES

United States and Canada

Stratagene
11011 North Torrey Pines Road
La Jolla, CA 92037
Telephone (858) 373-6300
Order Toll Free (800) 424-5444
Technical Services (800) 894-1304
Internet tech_services@stratagene.com
World Wide Web www.stratagene.com

Stratagene European Contacts

Location	Telephone	Fax	Technical Services
Austria	0800 292 499	0800 292 496	0800 292 498
Belgium	00800 7000 7000	00800 7001 7001	00800 7400 7400
	0800 15775	0800 15740	0800 15720
France	00800 7000 7000	00800 7001 7001	00800 7400 7400
	0800 919 288	0800 919 287	0800 919 289
Germany	00800 7000 7000	00800 7001 7001	00800 7400 7400
	0800 182 8232	0800 182 8231	0800 182 8234
Netherlands	00800 7000 7000	00800 7001 7001	00800 7400 7400
	0800 023 0446	+31 (0)20 312 5700	0800 023 0448
Switzerland	00800 7000 7000	00800 7001 7001	00800 7400 7400
	0800 563 080	0800 563 082	0800 563 081
United Kingdom	00800 7000 7000	00800 7001 7001	00800 7400 7400
	0800 917 3282	0800 917 3283	0800 917 3281

All Other Countries

Please contact your local distributor. A complete list of distributors is available at *www.stratagene.com*.

Taq Extender™ PCR Additive

CONTENTS

Taq Extender™ PCR Additive

MATERIALS PROVIDED

Materials provided	Quantity
Taq Extender™ PCR additive (5 U/µl)	200 µl
Taq Extender™ 10× reaction buffer[a]	1 ml

[a] See *Preparation of Reagents*.

STORAGE CONDITIONS

All components: –20°C

ADDITIONAL MATERIALS REQUIRED

Taq DNA polymerase

Revision #046001b

INTRODUCTION

Taq Extender™ PCR additive is a polymerase enhancer that improves the reliability and yield of conventional polymerase chain reaction (PCR) amplifications. This additive increases the efficiency at which *Taq* DNA polymerase performs extension reactions on specific DNA segments in each cycle of PCR, thus resulting in a greater percentage of the extension reactions reaching completion. In addition, *Taq* Extender PCR additive improves the PCR amplification of difficult templates and increases the reliability and yield of many PCR targets up to 10 kb in length.[1] The easy-to-use *Taq* Extender PCR additive is simply added to amplification reactions in an amount equal to that of *Taq* DNA polymerase, the standard *Taq* 10× reaction buffer is replaced with an optimized *Taq* Extender 10× reaction buffer and cycling is performed using standard PCR conditions.

PROTOCOL

1. Prepare the amplification reaction using a standard PCR protocol and implement the modifications to the standard protocol as described in step 2. Determine the standard reaction parameters, such as the deoxynucleotide (dNTP) concentration, the amount of *Taq* DNA polymerase required and the cycling conditions (see step 3).

2. Modify the standard PCR protocol as outlined below.

   Notes *Modifications of primer length or cycling temperatures are not necessary.*

   Use of thin-wall microcentrifuge tubes is highly recommended, but not critical, to improve the heat transfer, which will further enhance the efficacy of the Taq *Extender PCR additive.*

   a. Substitute the standard *Taq* 10× reaction buffer with the *Taq* Extender 10× reaction buffer.

   b. Add an equal number of units of *Taq* Extender PCR additive and *Taq* DNA polymerase to the amplification reaction [i.e., add 1 µl of *Taq* Extender PCR additive (5 U/µl) to an amplification reaction requiring 1 µl of *Taq* DNA polymerase (5 U/µl)]. Use a minimum of 1 U each of *Taq* DNA polymerase and *Taq* Extender PCR additive per kilobase to be amplified.

3. Perform the amplification reaction using standard cycling conditions, allowing at least 30–60 seconds of extension time for each kilobase to be amplified.[2]

TROUBLESHOOTING

Observation	Solution(s)
Low yield	Minor lot-to-lot variations in the concentration of *Taq* DNA polymerases from various manufacturers may affect PCR product yields and may necessitate the use of greater or lesser amounts of *Taq* Extender PCR additive per amplification reaction in order to achieve optimal PCR results
	Choice of the *Taq* Extender 10× reaction buffer may also affect PCR product yields. If the *Taq* Extender 10× reaction buffer provided with this kit does not appear to be optimized for your particular amplification system, further buffer optimization can be facilitated using Stratagene's Opti-Prime™ PCR optimization kit
	Insufficient amounts of *Taq* DNA polymerase or inadequate extension times may also contribute to poor PCR product yields. Optimal PCR product yields are obtained by using a minimum of 1 U of *Taq* DNA polymerase per kilobase to be amplified and by allowing at least 30–60 seconds of extension time for each kilobase to be amplified[2]
Artifactual smears	PCR smearing may be due primarily to minor contaminants of the *Taq* DNA polymerase. Two possible solutions are to use a different lot number of the enzyme or to reduce the extension time

PREPARATION OF REAGENTS

Taq Extender™ 10× Reaction Buffer

200 mM Tris-HCl (pH 8.8)
100 mM KCl
100 mM $(NH_4)_2SO_4$
20 mM $MgSO_4$
1% Triton® X-100
1 mg/ml nuclease-free bovine serum albumin (BSA)

REFERENCES

1. Nielson, K., Schoettlin, W., Bauer, J. C. and Mathur, E. (1994) *Strategies* 7(2):27.
2. Innis, M. A., Myambo, K. B., Gelfand, D. H. and Brow, M. A. (1988) *Proc Natl Acad Sci U S A* 85(24):9436-40.

ENDNOTES