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AccuPower® PCR Master Mix is a new, powerful, and ready-to-use PCR reagent optimized for more accurate PCR amplifications

I. Description

The *AccuPower®* PCR Master Mix contains DNA polymerase, dNTPs and reaction buffer in a 2xMaster mixed solution format. Patented chemical stabilizer maintains the activity of the 2xMaster mixture for over 1 years in the freezer.

II. Storage

store at -20°C

III. Application

- Routine PCR
- Primer extension
- TA cloning
- Gene sequencing

IV. Advantages

- Speed : Substantial reduction in PCR setup time.
- Stability : *AccuPower®* PCR Master Mix is the powerful technology for convenient and easy to perform DNA amplification. It contains DNA polymerase, dNTPs, a tracking dye and reaction buffer in a master mixed solution format. The patented chemical stabilizer of this product enables to maintain the activity of pre mixture for over 1 years in the freezer. It ensures superior amplification efficiency with experiment stability and uniform activity of polymerase in the process of PCR.
- Reproducibility and Yield : Batch manufacturing under strict ISO 9001 quality control conditions guarantees reproducibility. Minimal handling during reaction set up provides improved accuracy.
- Simplicity : Each product line contains an application specific enzyme in an easy to re-suspended, lyophilized 2xmaster mix of dNTPs, reaction buffer, a tracking dye, and a stabilizer. The 2xmaster mix is ready to use, thus offering virtually no reaction set up time.

V. Notice to Purchaser

This enzyme is specifically optimized for increasing base incorporation rate by inactivating 5'→3' exonuclease activity. Therefore, this is not recommended to use for Real Time PCR using Taqman® probe. Bioneer corporation reserves the right to make corrections, modifications, improvements and other changes to its products, services, specifications or product descriptions at any time without notice. All information provided here is subject to change without notice.

VI. Contents

Reaction size	20 µl reaction (2xMasterMix 10 µl)	50 µl reaction (2xMasterMix 25 µl)
Top DNA polymerase	1 U	2.5 U
dNTP(dATP, dCTP, dGTP, dTTP)	Each 250 µM	Each 250 µM
Reaction Buffer, with 1.5mM MgCl ₂	1X	1X
Stabilizer and tracking dye	O	O

VII. Experimental Protocol

The following protocol is suggested as a general guideline when using *AccuPower®* PCR Master Mix in any PCR amplification.

- 1) Add template DNA and primers into 2xPCR Master Mix tubes.
- 2) Add distilled water into mixture tubes to a total volume of 20 µl or 50 µl. Do not calculate the dried pellet.

Ex) Reaction Mix

Components	20 µl reaction volume	50 µl reaction volume
2xPCR Master Mix	10 µl	25 µl
Template DNA	Variable(1~5 µl)	Variable(1~15 µl)
Forward Primer (10 pmole/ µl)	0.5 µl ~2 µl	1 µl ~5 µl
Reverse Primer (10 pmole/ µl)	0.5 µl ~2 µl	1 µl ~5 µl
D.W.	Variable	Variable
Total volume	20 µl	50 µl

► Note 1 : Amount of template

Template DNA	Amount of template	
	20 µl reaction	50 µl reaction
Bacteriophage λ, Plasmid DNA	100 fg~200 ng	100 fg~500 ng
Total Genomic DNA	1 ng~500 ng	1 ng~1 ug

- 3) Mixture by flick with your finger or pipetting, and briefly spin down.
- 4) (Option) If necessary, overlay mineral oil. This step is unnecessary when using a thermal cycler with top heating.
- 5) Perform the reaction under the following conditions.

• In case of routine PCR

Step	Temperature	Time	Cycles
Pre-Denaturation	95°C	5 min	1 cycle
Denaturation	95°C	20 sec	25~35 cycles
Annealing	45~65°C	20 sec	
Extension	72°C	30 sec ~ 1 min/kb	
Final Extension	72°C	Optional. Normally 3~5 min	1 cycle

• In case Primer's T_m value is more than 65°C or PCR Product size is more than 5 kb.

Step	Temperature	Time	Cycles
Pre-Denaturation	95°C	5 min	1 cycle
Denaturation	95°C	20 sec	30~35 cycles
Anneal/Extension	68°C	1 min/kb	
Final Extension	72°C	Optional. Normally 3~5 min	1 cycle

- 6) Maintain the reaction at 4°C~8°C after cycling. The samples can be stored at -20°C until use.
- 7) Load samples on agarose gel without adding a loading-dye mixture and perform electrophoresis.

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VIII. Experimental Data

1) Sensitivity.

• PCR Condition

▶ Bioneer's AccuPower® PCR PreMix

Temperature	Time	Cycles
95°C	5 min	1 cycle
95°C	20 sec	35 cycles
55°C	20 sec	
72°C	30 sec	
72°C	3 min	1 cycle

▶ Others company : Reactions were performed following suppliers' recommendations

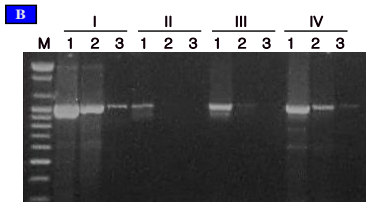
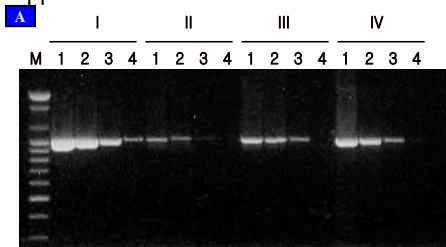


Figure 1. Comparison of sensitivity test for PCR Master Mix and other companies' products using serial diluted human genomic DNA. Amplification of A the human Insulin receptor gene and B the DNA cross-link repair 1A gene.

M : 100 bp DNA Ladder (Bioneer Cat. No : D-1030)
Lane 1 : Human DNA 10 ng Lane 2 : Human DNA 1 ng
Lane 3 : Human DNA 100 pg Lane 4 : Human DNA 10 pg
I : AccuPower® PCR Master Mix (Bioneer)
II : A company Taq DNA polymerase
III : B company Taq DNA polymerase
IV : C company PCR PreMix

2) Long kb.

• PCR Condition

▶ Bioneer's AccuPower® PCR Master Mix

- In case of Lane 1, 2 (3 step)

Temperature	Time	Cycles
95°C	5 min	1 cycle
95°C	20 sec	30 cycles
55°C	20 sec	
72°C	30 sec	
72°C	3 min	1 cycle

- In case of Lane 3, 4 (2 step)

Temperature	Time	Cycles
95°C	5 min	1 cycle
95°C	20 sec	30 cycles
68°C	4 min	
72°C	3 min	1 cycle

▶ Others company : Reactions were performed following suppliers' recommendations

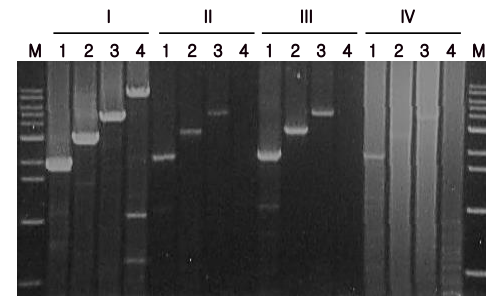


Figure 5. Comparison of long kb test for AccuPower® PCR PreMix and other companies' products using human genomic DNA.

Lane 1: 2 kb fragment of human TP53 gene
Lane 2: 3 kb fragment of human TP53 gene
Lane 3: 4.5 kb fragment of human DCLRE1A gene
Lane 4: 8 kb fragment of human HBB region
M: 1 kb DNA ladder (Bioneer Cat. No : D-1040)
I : AccuPower® PCR Master Mix (Bioneer)
II : A company Taq DNA polymerase
III : B company Taq DNA polymerase
IV : C company PCR PreMix

IX. Ordering Information

Tube type	Reaction	Cat.No.	Description
0.2 ml Tube	20 µl	K-2012	0.2 ml thin-wall 8-strip tubes with attached cap / 96 tubes
		K-2016	0.2 ml thin-wall 8-strip tubes with attached cap / 480 tubes
		K-2036	0.2 ml thin-wall 8-strip tubes with attached cap / 96 tubes / Negative Dye
	50 µl	K-2037	0.2 ml thin-wall 8-strip tubes with attached cap / 480 tubes / Negative Dye
		K-2013	0.2 ml thin-wall 8-strip tubes with attached cap / 96 tubes
		K-2017	0.2 ml thin-wall 8-strip tubes with attached cap / 480 tubes
0.5 ml Tube	50 µl	K-2011	0.5 ml thin-wall tubes with attached cap / 100 tubes
96-well	10 µl	K-2260-1	thin-wall 96-well flat plate
		K-2260-2	thin-wall 96-well full-skirted plate
		K-2260-3	thin-wall 96-well semi-skirted plate
	20 µl	K-2260-4	thin-wall 96-well flat plate
		K-2260-5	thin-wall 96-well full-skirted plate
		K-2260-6	thin-wall 96-well semi-skirted plate
384-well	5 µl	K-2080-1	thin-wall 384-well full-skirted plate
	10 µl	K-2080-2	thin-wall 384-well full-skirted plate
	20 µl	K-2080-3	thin-wall 384-well full-skirted plate
Master Mix	-	K-2018	1 ml of 2 X Master mix solution
		K-2018-1	10 ml of 2 X Master mix solution