5x PCR-Mastermix "ready-to-load"

Data sheet

Article no BS 91.731.1000	**250 Reactions x 20 µl**
Article no BS 91.731.1000 x 3	**750 Reactions x 20 µl**
Article no BS 91.731.1000 x 5	**1250 Reactions x 20 µl**

(For research and *in vitro* use only)

Batch-Nr.:	BS.25FMG0630
Expiry:	02-2020
Appearance:	clear liquid
Colour:	green

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Description

Bio&SELL 5x PCR Master Mix "ready-to-load" is a premixed ready-to-use solution containing all reagents required for PCR (except template, primer and water), additional compound needed for direct loading onto agarose gel and two tracking dyes (blue and yellow) that allow to monitor progress during electrophoresis.

Composition

- Bio&SELL Taq-DNA-Polymerase
- 5x reaction buffer B
  (400 mM Tris-HCl pH 9.4 – 9.5 at 25°C, 100 mM $(NH_4)_2SO_4$, 0.1% w/v Tween-20)
- 12.5 mM $MgCl_2$ (= 2,5 mM in 1x Solution)
- 1 mM of each dATP, dGTP, dCCT and dTTP
  (= 200 µM per dNTP in 1x solution)
- Blue and yellow dye and additional compounds for direct loading onto agarose gels.

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Recommended Protocol

The components should be pipetted according to this protocol (here for 50 µl final volume).

Component	Volume	Final concentration
5x PCR-Mastermix	10 µl	1x
Primer 1	x µl	0,4 µM
Primer 2	x µl	0,4 µM
Template-DNA	x µl	5 – 100 ng
Water	Add water up to 50 µl	

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Technical Data

Source: Based on the thermostable DNA polymerase, isolated from the thermophilic bacterium Thermus aquaticus. The enzyme used here is recombinantly expressed in E. coli.

Analysis conditions: 25 mM Tris•HCl (pH 9,0 at 25°C), 50 mM KCl, 2 mM MgCl2, 0,1mg/ml gelatine, 200 µM of each dATP, dGTP, dTTP, 100 µM [α32-P]dCTP (0,05 µCi/nmol), 12,5 µg activated salmon sperm DNA.

Unit definition: One unit is the amount of enzyme required to catalyze the incorproation of 10 nmol of dNTPs into acid-insoluble form in 30 min at 74ºC.

Storage and dilution buffer: 50% glycerole (v/v), 20 mM Tris•HCl (pH 8.7 at 25ºC), 100 mM KCl, 0.1 mM EDTA and stabilizers.

Enzyme activities: Bio&SELL Taq DNA polymerase is a highly processive 5' à 3'-DNA polymerase with 5' à 3'-exonuklease activity. A 3' à 5'-Exonuklease activity lacks completely. Additionally, the enzyme adds single nucleotides (almost exclusively adenosines) to the 3´-ends of the DNA, so that TA cloning is possible without further modifications.

Application & Quality control: Primer extension reaction: the enzyme is free of nicking and primer extension activities as well as of exonucleases and unspecific endonucleases. SDS/PAGE: 95 kD-band, purity >98%. Activity and stability tested via PCR. The error rate per nucleotide per cycle is ~8.3x10-5; the accuracy ~ 1.2x104. Estimated half life at 95ºC: 90 min.

Shipment Room temperature; Storage at -20°C
(Storage at room temperature for up to one month is possible without loss of activity, storage at 2 – 8°C is possible for up to 6 months.)

Note: *Bio&SELL* 5x PCR Master Mix "Ready-to-Load" is optimized for PCR reactions and detection of the PCR products by agarose electrophoresis. The blue dye migrates in an 1% agarose gel with the same speed as 3,5 – 4,5 kb DNA fragments, the yellow dye with the same speed as the primers (< 35 – 45 bp).

Attention: *Bio&SELL* 5x PCR Master Mix "Ready-to-Load" contains Tween-20. If a detergent-free buffer system is required, please contact us. Phone: + 49 (0) 9128 – 724 32 32
Bio&SELL 5x PCR-Master Mix "Ready-to-Load" is not recommended for applications with spectrophotometrical analyses (detection of absorptions or fluorescence), as the two dyes could interfere with those applications.

Safety warnings and precautions: This product and its components should be handled only by persons trained in laboratory techniques. It is advisable that suitable protective clothing, such as laboratory overalls, safety glasses and gloves, are worn. Care should be taken to avoid contact with skin or eyes. In case of contact wash immediately with water.

Note: Some applications in which this product is used may require a license which is not provided by the purchase of this product. Users should obtain the license if required.

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