

Q-Extract DNA Extraction PCR Kit

With Taq DNA Polymerase 2x Master Mix RED
1.5 mM MgCl₂ final concentration



Cat. No.: A570004

A570004

500 Reactions

	Q-Extract DNA Extraction Solution	Taq DNA Polymerase 2x Master Mix RED, 1.5 mM MgCl ₂
ID No.	4900100	5200300
Cap colour	Clear	Red
Content	5 x 10 ml	5 x 1.25 ml

MADE IN DENMARK

Product description

Q-Extract DNA Extraction PCR Kit consists of Q-Extract DNA extraction solution and Taq DNA Polymerase 2x Master Mix RED, which is required for the subsequent PCR reaction.

The Q-Extract DNA Extraction Solution is designed for rapid and efficient extraction of PCR-ready DNA from many different sample types e.g. mouse tails or ears, saliva, bacteria and various mammalian tissues. The non-toxic Q-Extract DNA Extraction Solution enables the extraction of DNA from tissues in just 8 minutes. The extraction protocol is divided into two simple heating steps which is directly followed by PCR using Taq DNA Polymerase 2x Master Mix RED. This method is ideal for PCR analysis, such as screening and genotyping.

The one-reagent DNA extraction set-up is easily scaled and can be conducted by robotic automation platforms. Depending on the sample size, the DNA extraction can be performed in PCR tubes or 1.5 ml tubes, using either a thermocycler or heating block.

Taq DNA Polymerase 2x Master Mix RED is a ready-to-use 2x reaction mix. Each PCR reaction requires 12.5 µl of the master mix. Simply add primers, DNA extract and water to a total reaction volume of 25 µl to successfully carry out PCR.

This kit combination allows for DNA extraction and amplification hereof in less than 1½ hour, as compared to ≥1 day with conventional protocols.

Composition of Q-Extract DNA Extraction Solution

- Optimized DNA extraction solution

Composition of Taq DNA Polymerase 2x Master Mix RED

- Tris-HCl pH 8.5, (NH₄)₂SO₄, 3 mM MgCl₂, 0.2% Tween® 20
- 0.4 mM of each dNTP
- Ampliqon Taq DNA polymerase
- Inert red dye and stabilizer

Recommended Storage and Stability of Kit Components

Q-Extract DNA Extraction Solution: Long term storage at -20 °C. Product expiry at -20 °C is stated on the label. Can be stored short term at +4 °C for up to 3 months. Q-Extract DNA Extraction Solution tolerates up to 20 freeze-thaw cycles. It is recommended to aliquot the Q-Extract into smaller volumes.

Taq DNA Polymerase 2x Master Mix RED: Long term storage at -20 °C. Product expiry at -20 °C is stated on the label. Can be stored at +4 °C for up to 6 months.

Quality Control

Each batch of Q-Extract DNA Extraction Solution is functionally tested.

Taq DNA Polymerase is functionally tested and tested for contaminating activities, with no traces of endonuclease activity, nicking activity or exonuclease activity.

Extraction Protocol

Preparation of DNA extraction should be performed in a separate area from that used for setting up the PCR reaction.

- Thaw Q-Extract DNA Extraction Solution. For the first time use, aliquot the Q-Extract DNA Extraction solution into smaller volumes. (Q-Extract DNA Extraction Solution has a cloudy appearance).
- Add your sample to a tube containing 100 µl Q-Extract DNA Extraction Solution. Recommended sample sizes are shown in Table 1.
- Vortex the tube containing the sample and the DNA extraction solution for 15 sec.
- Transfer the tube to a heat block or a thermal cycler and incubate for
 - 65 °C for 6 min
 - 98 °C for 2 min.
 - 4 °C (or cool down on ice)

The DNA extract is now ready for PCR

DNA extracts are stable at -20 °C for one week or long term at -80 °C for longer periods.

- Mix the DNA extract with Taq DNA Polymerase 2x Master Mix RED. See PCR protocol and table 2.

Table 1. Sample sizes

Sample	Reaction volume 100 µl	Reaction volume 500 µl
Tissue*	0,5 – 10 mg	10 – 50 mg
<i>E. coli</i>	1 colony	1 colony
Saliva	-	~ 200 µl

* Examples of tested tissues include mouse tail, mouse organs and chicken breast.

PCR Protocol

This protocol serves as a guideline to ensure optimal PCR results when using Taq DNA Polymerase 2x Master Mix RED. Optimal reaction conditions such as incubation times, temperatures, and amount of template DNA may vary and must be determined individually.

1. Thaw Taq 2x Master Mix RED and primers.
It is important to thaw the solutions completely and mix thoroughly before use to avoid localized concentrations of salts.
Keep all components on ice.
2. Prepare a reaction mix. Table 2 shows the reaction set up for a final volume of 25 µL. If desired, the reaction size may be scaled up or down.

Table 2. Reaction components (reaction mix and template DNA)

Component	Vol./reaction*	Final concentration*
Taq 2x Master Mix	12.5 µl	1x
25 mM MgCl ₂	Optional	1.5 mM (1.5 – 4.5 mM)
Primer A (10 µM)	0.5 µl (0.25 – 2.5 µl)	0.2 µM (0.1 – 1.0 µM)
Primer B (10 µM)	0.5 µl (0.25 – 2.5 µl)	0.2 µM (0.1 – 1.0 µM)
PCR-grade H ₂ O	X µl	-
DNA Extract**	2 - 5 µl	Variable
TOTAL volume	25 µl	-

* Suggested starting conditions; theoretically used conditions in brackets

** If the PCR yields are poor or one experience no bands, it might help to dilute the DNA extract 1:10

3. Mix gently.
4. Add extracted DNA to the individual tubes containing the reaction mix.
5. Program the thermal cycler according to the manufacturer's instructions. See table 3 for an example.
For maximum yield and specificity, temperatures and cycling times should be optimized for each new template target or primer pair.
6. Place the tubes in the thermal cycler and start the reaction.
7. At the end of the run, simply load a portion of the reaction product (e.g. 10 µl) onto an agarose gel for analysis.

Table 3. Three-step PCR program

Cycles	Duration of cycle	Temperature
1	2 – 5 minutes	95 °C
25 - 35	20 – 30 seconds 20 – 40 seconds 30 seconds	95 °C 50 – 65 °C 72 °C
1	5 minutes	72 °C

Two-step PCR program

Fast 2-step PCR protocols are available using this link:
<https://ampliqon.com/en/pcr-technology/application-notes/>

Notes:

- The final MgCl₂ concentration of this 2x Taq Master Mix RED is 1.5 mM. In some applications, more than 1.5 mM MgCl₂ is required for best results. Use 25 mM to adjust the Mg²⁺ concentration according to table 4.

Table 4. Additional volume (µl) of MgCl₂ per 25 µl reaction:

Final MgCl ₂ conc. in reaction (mM)	1.5	2.0	2.5	3.0	3.5	4.0	4.5
Volume of 25 mM MgCl ₂	0	0.5	1	1.5	2	2.5	3

Related Products

Q-Extract DNA Extraction PCR Kit	Cat. No.
500 DNA extractions + PCR reactions	A570001

Q-Extract DNA Extraction Solution	Cat. No.
100 DNA extractions	A560001
500 DNA extractions	A560004

Taq Master Mixes (500 x 50 µl reactions) *	Cat. No.
2x Master Mix, 1.5 mM MgCl ₂ final concentration	A140303
2x Taq OptiMix CLEAR, 1.5 mM MgCl ₂ final concentration	A370503
2x Master Mix RED, 1.5 mM MgCl ₂ final concentration	A180303

TEMPase Hot Start Master Mixes (500 x 50 µl reactions)*	Cat. No.
2x Master Mix A**, 1.5 mM MgCl ₂ final concentration	A230303
2x Master Mix A**BLUE, 1.5 mM MgCl ₂ final concentration	A290403

*Master mixes available also in 1.1x variants as well as 2 mM MgCl₂ variants, **Mix A is Ammonium Buffer based, also available as Mix C based on Combination Buffer.

Special TEMPase Master Mixes (500 x 50 µl reactions)	Cat. No.
Multiplex 2x Master Mix, 3 mM MgCl ₂ final concentration	A260303
GC TEMPase 2x Master Mix I – for GC-rich templates	A331703
GC TEMPase 2x Master Mix II – for GC-rich templates	A332703

Taq DNA Polymerase (500 units) *	Cat. No.
Taq DNA Polymerase 5 U/µl	A110003
• with 10x Ammonium Buffer	A111103

*Available in kits including one or two buffers (Ammonium Buffer, Standard Buffer or Combination Buffer). All kits include extra 25 mM MgCl₂

Hot Start DNA Polymerase (500 units) *	Cat. No.
TEMPase Hot Start DNA Polymerase, 5 U/µl	A220003
• with 10x Ammonium Buffer	A221103

*Available in kits including one or two buffers (Ammonium Buffer, Standard Buffer or Combination Buffer). All kits include extra 25 mM MgCl₂

Buffers for DNA polymerases *	Cat. No.
10x Ammonium Buffer, 3 x 1.5 ml	A301103
10x Standard Buffer, 3 x 1.5 ml	A302103
10x Combination Buffer, 3 x 1.5 ml	A303103
5x PCR Buffer RED, 6 x 1,5 ml **	A301810
PCR Grade Water, 6 x 5 ml	A360056

*Ammonium Buffer, Standard Buffer and Combination Buffer are also available as Mg²⁺ free buffers, detergent free buffers and Mg²⁺ and detergent free buffers.
**For direct gel loading and visualisation.

Reagents for *in vitro* laboratory use only.

Other product sizes, combinations and customized solutions are available. Please look at www.ampliqon.com or ask for our complete product list for PCR Enzymes. For customized solutions please contact us.

Made in Denmark

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