PCR Mycoplasma Detection Kit

Catalogue No.:abx298017

PCR Mycoplasma Detection Kit is designed to detect the presence of mycoplasma contamination by PCR in biological samples, such as cell culture supernatants, which can be tested directly without DNA extraction. Highly specific primers have been designed to amplify a fragment of 16S rRNA coding DNA that is conserved across all commonly-known mycoplasma species. This kit is able to detect as low as 20 copies of the mycoplasma genome and only detects mycoplasma DNA, not eukaryotic or bacterial DNA. The kit includes an optimized supermix and primer, ultrapure water and positive control template. The kit provides a very easy to use, simple, fast (within 2 hours), specific and sensitive PCR-based mycoplasma detection method.

Kit components:

- 2X PCR Myco SuperMix: 1 ml
- Myco Primer Mix: 40 µl
- Myco Positive Control Template: 40 µl
- Myco-Free Water: 1 ml

Target:	PCR Mycoplasma Detection Kit
Storage:	Store at -20 °C for up to two years.
Note:	THIS PRODUCT IS FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC, THERAPEUTIC OR COSMETIC PROCEDURES. NOT FOR HUMAN OR ANIMAL CONSUMPTION.

Abbexa LTD, Cambridge, UK · Phone: +44 (0) 1223 755950 · Fax: +44 (0) 1223 755951
Abbexa LLC, Houston, TX USA · Phone: +1 832 327 7413
Abbexa BV, Leiden, NL
Website: www.abbexa.com · Email: info@abbexa.com

Directions for use:

Sample Preparation:

• **Adherent cells:** Cells should be cultured for 2-3 days with ~80% confluence. Transfer 40 µl of cell culture supernatant to a PCR tube and incubate at 95 °C for 10 minutes in a thermocyler.

• **Suspension cells:** Cells should be cultured for 2-3 days with a density of ~10^6 cells/ml. Transfer the cell culture to a microcentrifuge tube and centrifuge at 2000 × g for 1 minute. Transfer 40 µl of cell culture supernatant to a PCR tube and incubate at 95 °C for 10 minutes in a thermocyler.

• **Serum:** Dilute serum 20-fold. Transfer 40 µl of diluted serum to a PCR tube and incubate at 95 °C for 10 minutes in a thermocyler.

Samples can be stored at -20 °C for 1 month before or after heat treatment. Cells that have been cultured for more than 4 days should be diluted 10-fold before being tested in PCR.

PCR Setup – Reaction Components:

Component	**Volume**
Template	2 µl
Myco Primer Mix	0.4 µl
2X Myco PCR SuperMix	10 µl
Myco-Free Water	7.6 µl
Total Volume	**20 µl**

The provided Myco-Free Water can be used as a negative control and Myco Positive Control Template as the positive control.

PCR Amplification – Thermal Cycling Conditions:

Number of Cycles	**Temperature**	**Time per Cycle**
1 cycle	94 °C	4 min
35 cycles	94 °C	30 seconds
	60 °C	30 seconds
	72 °C	30 seconds
1 cycle	72 °C	5 min

Results and Analysis:

1. **Agarose gel electrophoresis:** Load 10 µl of PCR product onto a 1.5% agarose gel and resolve by electrophoresis.
2. **Results Interpretation:** Compare with the positive and negative control. The size of the positive band is about 350 bp.

Notes:

• To avoid mycoplasma contamination, the PCR reactions should be carried out in a designated PCR area and all reagents should be kept on ice during set-up. Face masks are recommended, as the oral cavity contains mycoplasma which may contaminate samples.
• Before use, completely thaw all components and mix well.
• Antibiotics such as penicillin, streptomycin, or serum in cell culture samples will not interfere with the assay.
• This product can be used together with abx098886 Mycoplasma Prevention Reagent and/or abx098884 and abx098885 Mycoplasma Elimination Reagents.

Abbexa LTD, Cambridge, UK · Phone: +44 (0) 1223 755950 · Fax: +44 (0) 1223 755951
Abbexa LLC, Houston, TX USA · Phone: +1 832 327 7413
Abbexa BV, Leiden, NL
Website: www.abbexa.com · Email: info@abbexa.com