



Mycoplasma PCR Detection Kit

Cat. No. G238

Store at -20°C.

Product Description

The **Mycoplasma PCR Detection Kit** offers **high specificity and sensitivity** to minimize false positives while ensuring coverage over 200 strains of *Mycoplasmas* for quick and reliable routine screening of cell cultures less than 1 hour. *Mycoplasmas* are highly undesirable, easily acquired and notorious for an elusive onset of infection. It can alter the infected cells at a molecular level leading to visible changes in cell morphology and growth characteristics. Timely detection of *Mycoplasmas* in cell cultures is recommended in order to deter wide-spread contamination and save on the costly efforts of elimination. The MasterMix contains gel loading dye, making it convenient to load for gel electrophoresis.

Product Component	Quantity	Part No.
BlasTaq™ 2X PCR MasterMix	100 rxn (1.25 ml)	P895-1
Primer Mix	100 µl	P238-2
Positive Control	250 µl	P238-3
Nuclease-Free H ₂ O	1.0 ml	P100

Protocol

1. Cells should remain in culture for at least 48-72 hours undisturbed prior to screening and is at least 80% confluence. Collect 2.5 µl directly from culture.
2. Mix individual components before use, and assemble reaction on ice.

Component	Volume
BlasTaq™ 2X PCR MasterMix	12.5 µl
Primer Mix	1 µl
Test Sample, Positive Control, or NTC (ddH ₂ O)	2.5 µl
Nuclease-Free H ₂ O	up to 25 µl

3. Gently mix the reaction and briefly centrifuge. Run thermocycling conditions for standard PCR:

Step	Temperature	Duration	Cycle(s)
Enzyme Activation	95°C	3 min	1
Denaturation	95°C	15 sec	30 - 40
Annealing	55°C	15 sec	
Extension	72°C	15 sec	
Final Extension	72°C	1 min	1
Holding	4°C	-	-

4. After PCR, maintain the reaction at 4°C or store at -20°C until use.
5. Analyze the amplification products by agarose gel electrophoresis.
6. Visualize by ethidium bromide or SafeView™ (Cat No. **G108**) staining.

General Notes

- PCR product approximately 500 bp in length indicates that the cell culture tested is contaminated with *Mycoplasma*. Note that the length of the PCR product will vary between 370-550 bp depending on the different *Mycoplasma* strains.
- Keep reaction mixture on ice prior to running the PCR, and start the PCR as soon as the reaction mixture is prepared.