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XpressMyco PCR Plus Detection Kit

Catalog No.: XMYCG-100

Description

The XpressMyco PCR Plus 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.

Kit Contents

Component	Quantity
2X PCR Master Mix	100 rxn (1.25 ml)
Primer Mix	100 µl
Positive Control	250 µl
Nuclease-Free H_2O	1.0 ml

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
2X PCR Master Mix	12.5 µl
Primer Mix	1 µl
Positive Control	2.5 µl
Nuclease-Free H_2O	Up to 25 µl

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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 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.

Important Information

These reagents are developed and sold for research purposes and in vitro use only. It is not intended for human or animal therapeutic or diagnostic purposes.