

Mycoplasma test kit

Cat. No. KC331-04/KC331-01

Store at -20°C.

Product Description

The **Mycoplasma test 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	KC331-01		KC331-04	
	Quantity	Part No.	Quantity	Part No.
BlasTaq™ 2X PCR MasterMix	100 rxn (1.25 ml)	P895- 1	20 rxn (250 ul)	P895- 4
Primer Mix	100 µl	P238-2	20 ul	P238-5
Positive Control	250 µl	P238-3	50 µl	P238-6
Nuclease- Free H ₂ O	1.0 ml	P100	200ul	P020

Protocol

- 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.
- 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

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

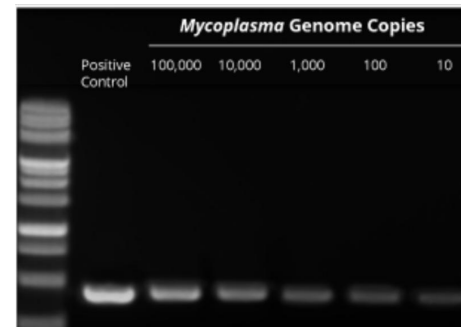
Step	Temperature	Duration	Cycle(s)
Enzyme Activation	95°C	3 min	1
Denaturation	95°C	15 sec	30 - 40
Annealing	55°C	15 sec	
Extension	72°C	15 sec	
Final Extension	72°C	1 min	1
Holding	4°C	-	-

- After PCR, maintain the reaction at 4°C or store at -20°C until use.
- Analyze the amplification products by agarose gel electrophoresis.
- Visualize by ethidium bromide or SafeView (Cat No. **K108**) 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.

Sensitive Detection



KEL's Mycoplasma PCR Detection Kit can detect samples containing as low as 10 genome copies of *Mycoplasma*.

KEL's Mycoplasma PCR Detection Kit (Cat. No. KC331-01) was used to detect samples containing ~10 to 100,000 genome copies of *Mycoplasma*.

Presence of *Mycoplasma* was confirmed as a 450 bp band.