molecular biology

genespin

info@genespin.com - www.genespin.com

cell cultures

NEW PRODUCTS

DATA SHEET

> 2X MycoPCR Master Mix GL green line

PCR test kit detection of mycoplasmas in cell cultures

2X MycoPCR Master Mix is a 2X premixed, ready-to-use solution (GeneSpin proprietary formulation) containing XtraTaq Pol, dNTPs, $MgCl_2$, specific mycoplasma primers set and stabilizers optimized for PCR-based mycoplasma detection in cell cultures. **2X MycoPCR Master Mix GL contain an internal fluorescent stain for DNA detection on Agarose gel directly after PCR amplification.** 2X MycoPCR Master Mix contains a combination of tartrazine and xylene dyes that allow gel loading and electrophoresis of the sample directly from the PCR tube, without further manipulation. The primer set allows detection of various mycoplasma species (e.g., M. fermentans, M. hyorhinis, M. arginini, M. orale, M. salivarium, M. hominis, M. pulmonis, M. arthritidis, M. bovis, M. pneumoniae, M. pirum and M. capricolum), as well as Acholeplasma and Spiroplasma species, with high sensitivity and specificity.

cat.no	*amount*	*note*
STS-MYCOP 150 GL	6x250ul	6x25 assay per kit with Positive Control
STS-MYCOP 50 GL	2x250ul	2x25 assay per kit with Positive Control
STS-MYCOP 25 GL	250ul	25 assay per kit with Positive Control

FOR RESEARCH USE ONLY

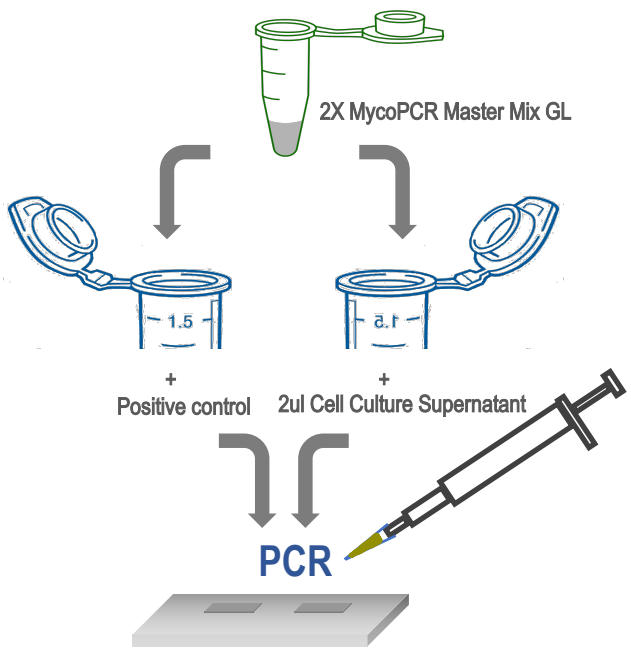
SHIPPING
Shipped in green ice.

STORAGE
Store at -20C°- Store dark.

SHELF LIFE
12 months

FORM
liquid green

CONCENTRATION
2X



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PROTOCOL

1. Transfer 100-200ul of cell culture supernatant into 1.5ml centrifuge tube. Incubate the supernatant at 95°C for 5 minutes.
2. Centrifuge at maximum speed for 5 minutes.
3. Use 2-5ul of the supernatant as PCR template.
   IMPORTANT! Before harvesting the supernatant from the cell culture, cells should cover approximately 90% of the growth surface! The supernatant may cause PCR inhibition in excessively dense cell cultures (>90%).

PCR assay set-up:

components	*test sample*	*control sample*
2X Myco PCR Master Mix GL	10ul	10ul
template from step 3	2-5ul	2ul
Water	8-5ul	8ul
	20ul reaction	

Cycling conditions:

Spin down the tubes/plate briefly to remove bubbles and place them into the cycler.

denaturation	95°C	5 min	1X
denaturation	95°C	60 sec	35X
annealing	58°C	90 sec	
extension	72°C	90 sec	
final elongation	95°C	5 min	1X
final step	4°C	∞	

DETECTION OF PCR PRODUCT

REMEMBER: **2X MycoPCR Master Mix GL contain an internal fluorescent stain for DNA detection on Agarose gel directly after PCR amplification.** This particular composition is able to avoid standard protocols for post- or pre- staining DNA on Agarose Gel with Ethidium Bromide (EtBr) or different dsDNA stains. The flouorescent used in GeneSpin PCR geen line regents has higher sensitivity than EtBr and has an easy, fast and robust staining procedure. Detection is possible by illuminating the Agarose Gel on a UV screen.
2X MycoPCR Master Mix a contains combination of tartrazine and xylene dyes that allow gel loading of the sample directly from the PCR tube and visualization of the sample during electrophoresis, without further manipulation.

4. For optimum separation we recommend using a 2% agarose gel with TAE or TBE buffer used for electrophoresis.
5. Load all PCR product volume (20ul) directly onto the gel and perform electrophoresis.
6. When the electrophoretic run is completed, lay the gel onto UV transilluminator to detect the expected band.
7. IF the test is POSITIVE, a DNA band of 750bp will appear.

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