

Myco-Sniff-Rapid™

Mycoplasma Luciferase Detection Kit

STORAGE

-20 °C in the dark for 1 year. Reconstituted Myco-Sniff-Rapid™ Reagent and Substrate can be stored at -80 °C for six months, at -20 °C for one month, or at 2-8 °C for one week. Repeated freezing and thawing up to 10 times will not affect the accuracy of the test results.

DESCRIPTION

Myco-Sniff-Rapid™ Mycoplasma Luciferase Detection Kit offers a quick (20 mins), simple and sensitive method for detecting mycoplasma contamination in cell cultures. This innovative kit utilizes the activity of mycoplasma metabolic enzymes from 209 mycoplasma species, including those commonly found contaminating cell cultures. Since this enzyme is not present in eukaryotic cells, the test is specific for mycoplasma. In the presence of mycoplasma, the enzymes react with the substrate to catalyze the conversion of ADP to ATP. Mycoplasma contamination can be detected by measuring the level of ATP in a sample before and after the addition of Myco-Sniff-Rapid™ substrate via a luciferase assay.

BENEFITS

- ▶ Rapid results in 20 minutes
- ▶ Highly sensitive
- ▶ Specific for detecting all common mycoplasma contaminant species
- ▶ No interference with cell culture media components
- ▶ Simple operation - add reagents directly to the culture supernatant

Components	093050401 (25 tests)	093050402 (50 tests)
Myco-Sniff-Rapid Reagent (lyophilized)	2 vials	4 vials
Myco-Sniff-Rapid Substrate (lyophilized)	2 vials	4 vials
Myco-Free Water	2 x 1.5 mL	4 x 1.5 mL

PROCEDURE

Materials Required, But Not Supplied

96-Well plate, Luminometer (tube luminometer, plate luminometer or multifunctional reader), Pipette, Pipette tips, Centrifuge

Component Reconstitution

Reconstitute one vial of lyophilized Myco-Sniff-Rapid Reagent and one vial of Myco-Sniff-Rapid Substrate by adding 700 µL Myco-Free water to each. Wait until rehydration is completed.

Collect Cell Culture

Culture the cells for at least 24 hours, collect the cell culture medium and centrifuge at 400 × g for 3 minutes. The supernatant should be tested immediately or stored at 2-8 °C for no more than one week. Avoid freezing and thawing the collected medium.

NOTE ▶ For optimal assay performance, cell confluency should reach 80% or higher.

Assay (avoid bright lighting)

- 1 Equilibrate the dissolved Myco-Sniff-Rapid Reagent, Myco-Sniff-Rapid Substrate and cell culture medium supernatant to room temperature.
- 2 Add 50 µL Myco-Sniff-Rapid Reagent and 50 µL cell culture medium supernatant to a 1.5 mL tube or 96-well plate and incubate at room temperature for 5-10 minutes.

NOTE ▶ Mix the samples gently with a pipette and be careful not to generate any large bubbles. Small bubbles on the edge of the tube or well should not impact assay result.

- 3 Place the tube or 96-well plate in a luminometer to measure the luminescent signal value (Reading A).
- 4 Add 50 µL of Myco-Sniff-Rapid Substrate to the tube or 96-well plate and incubate at room temperature for 10-15 minutes.

NOTE ▶ Mix the samples gently with a pipette and be careful not to generate any large bubbles. Small bubbles on the edge of the tube or well should not impact assay results.

- 5 Place the tube or 96-well plate in a luminometer to measure the luminescent signal value (Reading B).
- 6 Data interpretation: The ratio of Reading B to Reading A is used to determine whether a cell culture is contaminated by mycoplasma.

Ratio (Reading B/Reading A)	Results
$B/A \geq 1$	Mycoplasma contamination
$B/A \leq 0.8$	Negative for mycoplasma
$0.8 < B/A < 1$	Samples should be retested after another 24-48 hours culture in quarantine. If the B/A ratio remains between 0.8 and 1 with no significant increase, the sample is negative for mycoplasma.



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