

Mycoplasma Products

Keep Mycoplasma Out and Cell Purity In
with Powerful Anti-Mycoplasma Products



Mycoplasma Products for Managing Mycoplasma Contaminations

MycoAlert™ PLUS Mycoplasma Detection Kit

For accurate, reliable and universal mycoplasma detection

MycoZap™ Mycoplasma Elimination Reagent

For successful elimination of mycoplasma with low cell toxicity

MycoZap™ Plus-CL and Plus-PR

For protection against a broad range of microbial contaminants such as Gram(+) and Gram(-) bacteria, fungi as well as mycoplasma

MycoZap™ Prophylactic

For prevention of mycoplasma contamination in combination with your antibiotic formula of choice

What Are Mycoplasma?

- Belong to the family Mollicutes including *Mycoplasma*, *Acholeplasma*, *Ureaplasma*, *Entomoplasma*, *Spiroplasma* and other species
- Smallest free-living, self-replicating organisms (size: 0.2 µm – 0.8 µm)
- Simple prokaryocytes, lacking a rigid cell wall (surrounded by a single plasma membrane)
- Usually attached to the external surface of the cell membrane
- Relying on their hosts for many nutrients as their biosynthetic capabilities are limited
- Over 180 recognized species
- Six species account for 95% of all mycoplasma infections in cell cultures (*M. orale*, *M. arginii*, *M. fermentans*, *M. salivarum*, *M. hyorhina* and *A. laidlawii*)
- Widespread in nature as parasites of humans, mammals, reptiles, insects and plants

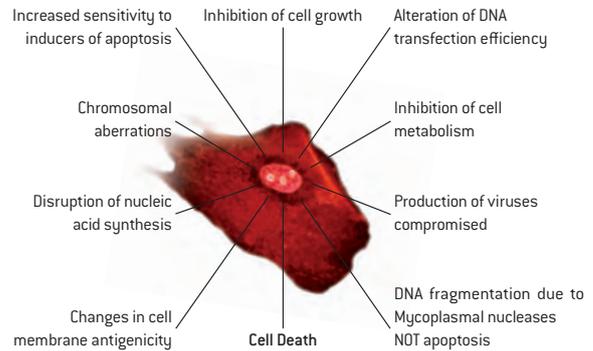
Typical Routes of Mycoplasma Infection in Cell Cultures

- Cross contamination from untested infected cells
- Aerosols created during pipetting
- Using the same bottle of medium for different cell types
- Handling more than one cell line in the hood at one time
- Contaminated materials
- Contaminated donor tissue (< 1%)
- Direct infection from the researcher

One of the most common contaminants present in cell culture laboratories are mycoplasma. A conservative estimate states that between 15–35% of all continuous cell cultures are contaminated with mycoplasma¹, some estimates are even higher (up to 80% in some countries)². Lonza provides a powerful product offering for reliable detection and successful elimination and prevention of mycoplasma contaminations.

How Does a Mycoplasma Contamination Affect Your Cells?

Mycoplasma grow slowly and do not kill the cells outright but affect various cellular parameters^{1,3,4,5,6,7,8,9,10,11} e.g.



Furthermore certain mycoplasma species are able to reduce tetrazolium salts causing aberrant results with tetrazolium assays, and so could mask any cytotoxic effects of compounds and cause shifts in IC₅₀ values.

How Insidious Are Mycoplasma?

Contaminations are very difficult to detect or prevent and the presence of mycoplasma can remain undiscovered for months:

- In contrast to bacteria, they do not cause visible changes in turbidity or pH
- Not visible under microscopy, even at very high concentrations > 10⁷ cfu/mL
- Most routine antibiotics used in cell culture are ineffective against mycoplasma
- They are not routinely removed by filtration

Thus, mycoplasma contaminations can seriously impact the reliability, reproducibility, and consistency of experimental results, representing a major problem for basic research as well as for the manufacturing of bioproducts. Standard testing for mycoplasma is an important quality control.

Detection: MycoAlert™ PLUS Mycoplasma Detection Kit – For Research Use

The MycoAlert™ Assay Kit is a selective biochemical test that exploits the activity of mycoplasmal enzymes which are found in all 6 of the main mycoplasma cell culture contaminants and the vast majority of 180 species of mycoplasma, but are not present in eukaryotic cells. The presence of these enzymes provides a rapid screening procedure allowing sensitive detection of low levels of contaminating mycoplasma in a test sample.

Benefits

- Results in <20 minutes by a simple 2-step assay
- Bioluminescence-based technology – no DNA extraction necessary
- NEW! MycoAlert™ PLUS Kit for use with less sensitive plate luminometers or multifunctional readers
- Convenient enzymatic assay control available for monitoring system performance

Applications

- Detects all common mycoplasma and acholeplasma contaminations
- Tested for 44 mollicute species so far
- Suited for cell culture screening in research environment
- Suited for testing of fresh media, supplements or water

Assay Principle

Viable mycoplasma are lysed and the enzymes react with the MycoAlert™ PLUS Substrate, catalysing the conversion of ADP to ATP. By measuring the level of ATP in a sample via a luciferase reaction, both before (read A) and after the addition of the MycoAlert™ PLUS Substrate (read B), a ratio can be obtained which is indicative of the presence or absence of mycoplasma. If mycoplasma are not present, the second reading shows no increase over the first, while reaction of mycoplasmal enzymes with their specific substrates in the MycoAlert™ PLUS Substrate leads to elevated ATP levels, resulting in more light output (Figure 1). The new next generation **MycoAlert™ PLUS Assay** generates a stronger light signal and thus provides broader compatibility with plate luminometers and multifunctional readers.

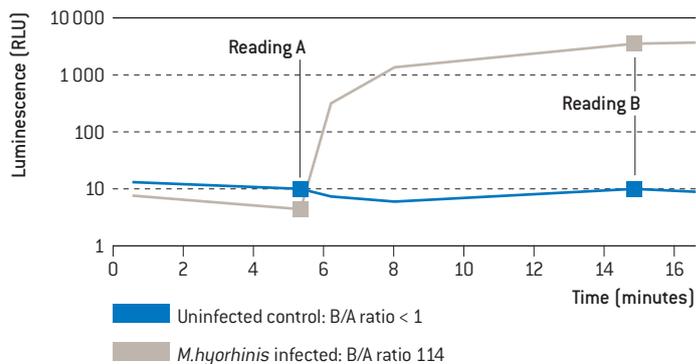


Figure 1. Kinetics of light emission for uninfected and infected cells using the MycoAlert™ PLUS Kit. The B/A ratio indicates the presence or absence of mycoplasma.

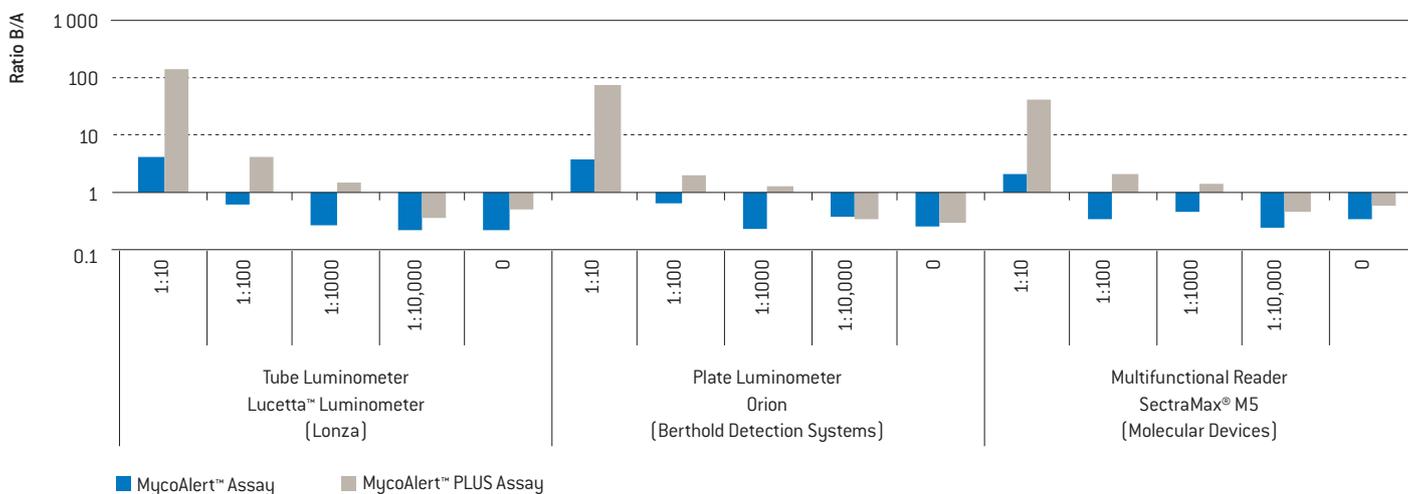


Figure 2. Performance of MycoAlert™ PLUS Assay compared to MycoAlert™ Assay. A dilution series of MycoAlert™ Assay Control demonstrates the increased sensitivity of various luminometer models when using MycoAlert™ PLUS Assay, compared to the first generation MycoAlert™ Assay.

Elimination and Prevention: MycoZap™ Product Line

MycoZap™ Mycoplasma Elimination Reagent

Mycoplasma cannot be removed by sterile filtration; and due to the lack of a cell wall, most routine antibiotics (e.g. penicillin) used in cell culture are ineffective against them.

Where contamination has occurred, and the sample absolutely cannot be discarded for certain reasons, the MycoZap™ Mycoplasma Elimination Reagent can clear detectable contamination in as few as four days (figure 4). The MycoZap™ Reagent has been optimized to eliminate mycoplasma with minimal toxic effects on the infected cells. It eliminates mycoplasma by using a combination of antibiotic and antimetabolic agents. This approach allows for a highly reliable elimination of mycoplasma that cannot be achieved by the use of antibiotics alone. The MycoZap™ Reagent can be used to eradicate mollicutes, including *Mycoplasma*, *Acholeplasma*, *Spiroplasma* and *Entomoplasma* species in cell cultures.

Universal and Thorough Elimination

- Eradicates *Mycoplasma*, *Acholeplasma*, *Spiroplasma* and *Entomoplasma*
- Efficient elimination of mollicutes by using a combination of antibiotic and antimetabolic agents

Effective But Mild

- Minimal toxic effects
- Suited for a broad range of cell cultures

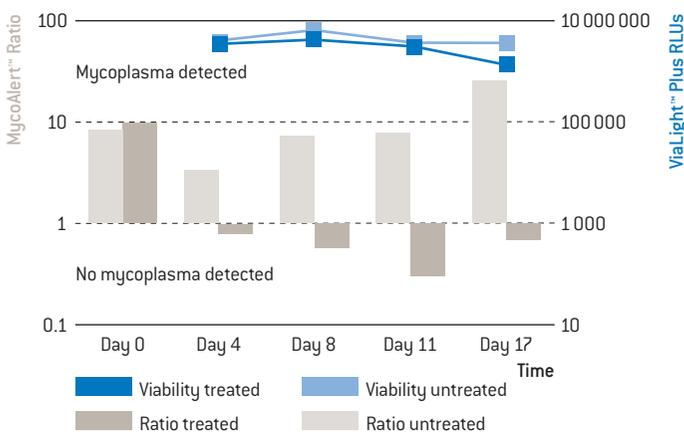


Figure 3. Effect of MycoZap™ Elimination Reagent on cell viability and mycoplasma removal. The MycoZap™ Reagent treatment eliminates mycoplasma in as few as 4 days with minimal impact on cell viability. Mycoplasma detection was performed with MycoAlert™ Assay and viability was determined by ViaLight™ Plus Assay.

MycoZap™ Antibiotics

MycoZap™ Antibiotics are extremely powerful combinations of innovative antibiotics for the protection of cell cultures from mycoplasma contamination. While MycoZap™ Prophylactic prevents mycoplasma contamination, MycoZap™ Plus offers protection against a broad range of common contaminants including mycoplasma. MycoZap™ Plus-CL is suited for protection of cell lines and MycoZap™ Plus-PR is optimized for gentle protection of primary cells.

| | Mycoplasma Only Solution | All-in-one Solutions | |
|--|---|----------------------|---------|
| | Prophylactic | Plus-CL | Plus-PR |
| Prevention against mycoplasma | ■ | ■ | ■ |
| Prevention against - Gram(+) bacteria - Gram(-) bacteria - Fungi - Yeast | No; but can be used in combination with other antibiotic formula of choice detection and prevention | ■ | ■ |
| Suited for primary cells | ■ | | ■ |
| Suited for cell lines | ■ | ■ | |

Ordering Information

| Cat. No. | Description | Size |
|-----------------------------|--|--------------|
| MycoAlert™ PLUS Kits | | |
| LT07-701 | MycoAlert™ PLUS Mycoplasma Detection Kit | 10 tests |
| LT07-703 | MycoAlert™ PLUS Mycoplasma Detection Kit | 30 tests |
| LT07-705 | MycoAlert™ PLUS Mycoplasma Detection Kit | 50 tests |
| LT07-710 | MycoAlert™ PLUS Mycoplasma Detection Kit | 100 tests |
| LT07-518 | MycoAlert™ Assay Control Set | 10 tests |
| MycoZap™ Products | | |
| LT07-818 | MycoZap™ Mycoplasma Elimination Reagent | 1 treatment |
| LT07-918 | MycoZap™ Mycoplasma Elimination Reagent | 5 treatments |
| VZA-2011 | MycoZap™ Plus-CL | 10 × 1 mL |
| VZA-2012 | MycoZap™ Plus-CL | 1 × 20 mL |
| VZA-2021 | MycoZap™ Plus-PR | 10 × 1 mL |
| VZA-2022 | MycoZap™ Plus-PR | 1 × 20 mL |
| VZA-2031 | MycoZap™ Prophylactic | 10 × 1 mL |
| VZA-2032 | MycoZap™ Prophylactic | 1 × 20 mL |

 For more information, please visit www.lonza.com/mycoplasma

References

- Drexler HG, Uphoff CC (2002) Mycoplasma contamination of cell cultures: Incidence, sources, effects, detection, elimination, prevention. *Cytotechnology* 39: 75–90
- Koshimizu K, Kotani H (1981). In: Procedures for the Isolation and Identification of Human, Animal and Plant Mycoplasmas (Nakamura, M., ed.), *Saikou, Tokyo*, 87–102.
- Gong H, Zölzer F, von Recklinghausen G, Rössler J, Breit S, Havers W, Fotsis T, Schweigerer L (1999) Arginine deiminase inhibits cell proliferation by arresting cell cycle and inducing apoptosis. *Biochem Biophys Res Comm* 261: 10–14.
- Ben-Menachem G, Mousa A, Brenner T, Pinto F, Zähringer U, Rottem S (2001) Choline deficiency induced by Mycoplasma fermentans enhances apoptosis of rat astrocytes. *FEMS Microbiol Letters* 201: 157–162
- McGarrity MF, Vanaman V, Sarama J (1984) Cytogenetic effects of mycoplasma infection of cell cultures: a review. *In Vitro* 20: 1–18
- Sokolova IA, Vaughan ATM, Khodarev NN (1998) Mycoplasma infection can sensitize host cells to apoptosis through contribution of apoptotic-like endonuclease(s). *Immunol Cell Biol* 76: 526–534
- Doersen CJ, Stanbridge EJ (1981) Effects of mycoplasma contamination on phenotypic expression of mitochondrial mutants in human cells. *Mol Cell Biol* 1: 321–329
- Stanbridge EJ (1971) Mycoplasmas and cell cultures. *Bacteriological Reviews* 35: 206–227
- Darin N, Kadhom N, Brière JJ, Chretien D, Bébéar CM, Rötig A, Munnich A, Rustin P (2003) Mitochondrial activities in human cultured skin fibroblasts contaminated by Mycoplasma hyorhinis. *BMC Biochem* 4: 15
- Rottem S (2003) Interaction of mycoplasmas with host cells. *Physiol Rev* 83: 417–432
- Miller CJ, Kassem HS, Pepper SD, Hey Y, Ward TH, Margison GP (2003) Mycoplasma infection significantly alters microarray gene expression profiles. *Biotechniques* 35: 812–814

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CD-BR022 06/17

