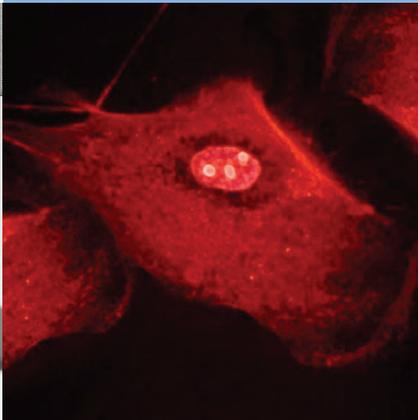


Mycoplasma Products

Keep Mycoplasma out and Cell Purity in
with Powerful Anti-Mycoplasma Products



Mycoplasma Products for Managing
Mycoplasma Contaminations



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.

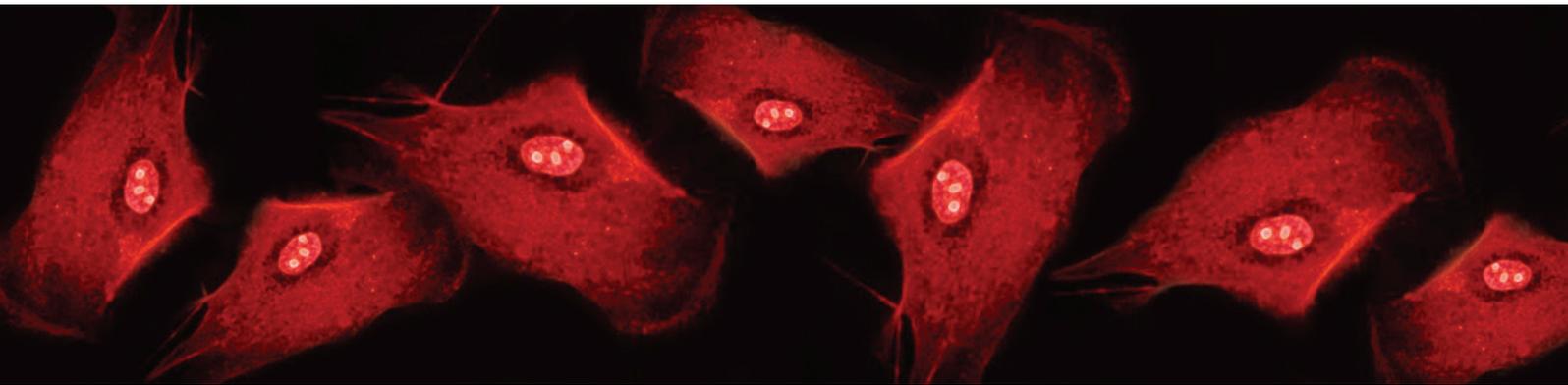
Lonza provides a powerful product offering for reliable detection and successful elimination and prevention of mycoplasma contaminations.

MycoAlert™ PLUS Mycoplasma Detection Kit

For accurate, reliable and universal mycoplasma detection

Lucetta™ Luminometer

For sensitive luminescence measurement and with a tailor-made mode for MycoAlert™ Mycoplasma Detection Kit



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

MycoZap™ Mycoplasma Elimination Reagent

For successful elimination of mycoplasma with low cell toxicity

MycoZap™ Prophylactic

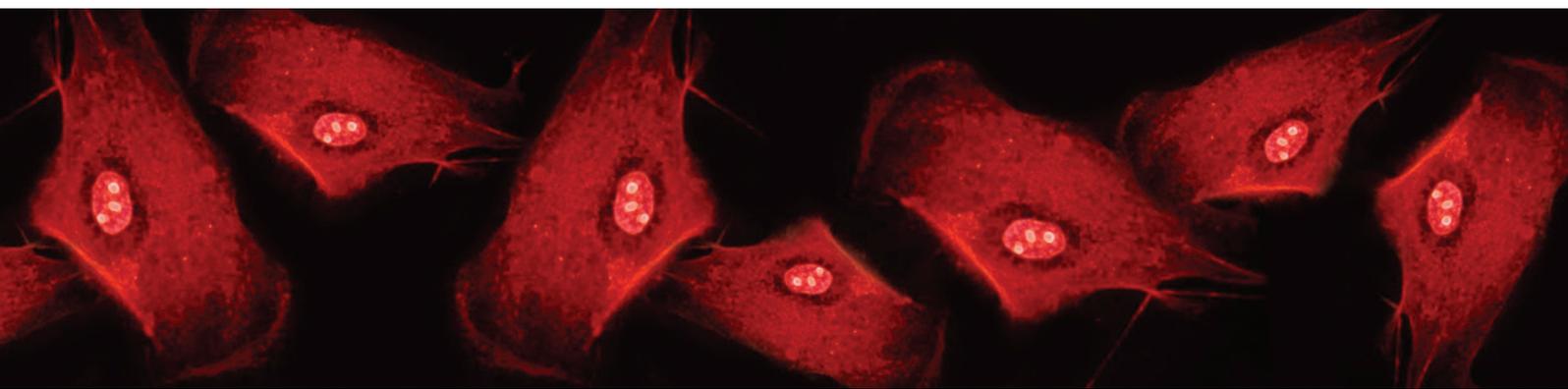
For prevention of mycoplasma contamination in combination with your antibiotic formula of choice

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

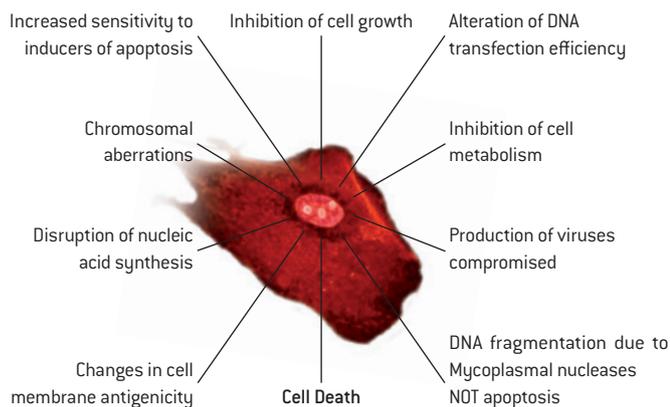
MycoZap™ Spray

For reliable disinfection of laboratory surfaces from mycoplasma contamination



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 IC_{50} 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 $> 10^7$ 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.

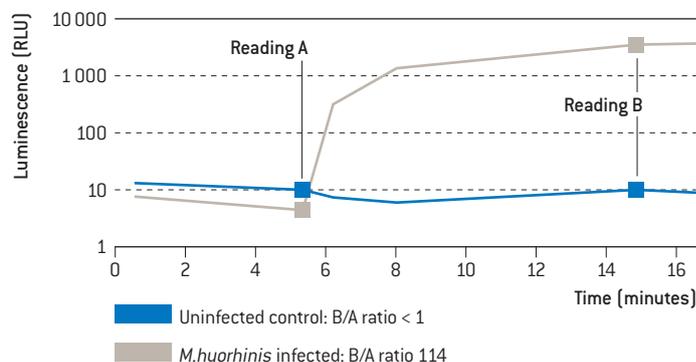


Figure 1
Kinetics of light emission for uninfected and infected cells using the MycoAlert™ PLUS Kit. The B/A ratio indicates the presence or absence of mycoplasma.

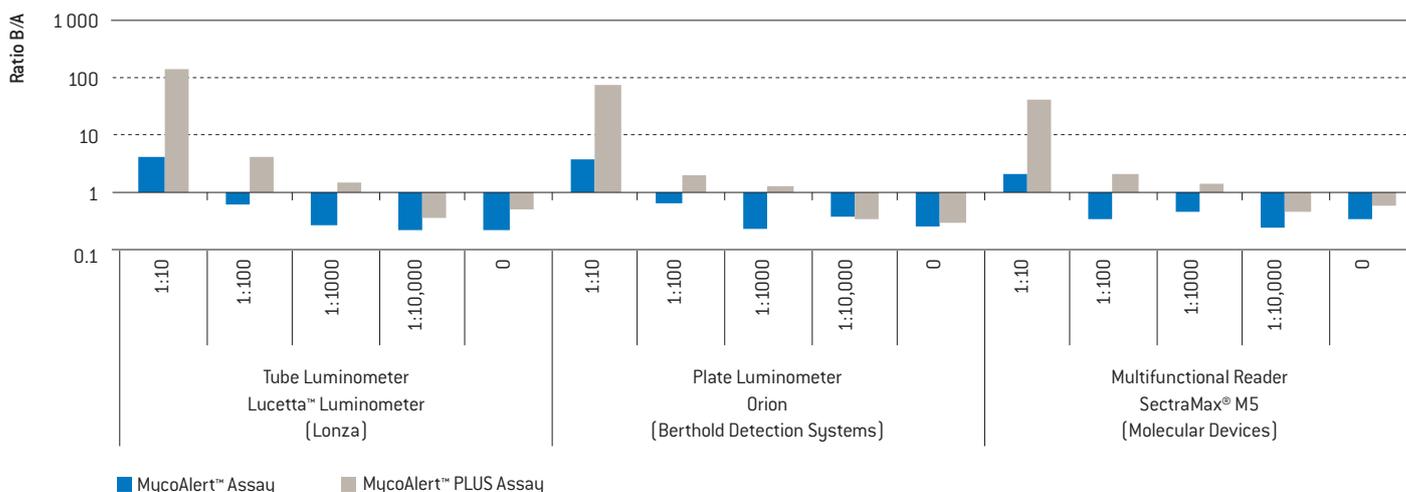


Figure 2
Performance of MycoAlert™ PLUS Assay compared to MycoAlert™ Assay. A dilution series of MycoAlert™ Assay Control demonstrates the increased sensitivity of various luminometer models when using MycoAlert™ PLUS Assay, compared to the first generation MycoAlert™ Assay.

Detection: Lucetta™ Luminometer Tailor-Made for MycoAlert™ Assay

The Lucetta™ Luminometer is a single sample tube luminometer for the detection of bioluminescence and chemiluminescence. The small, portable instrument is suited for all measurements of glow luminescence. It is designed with a tailor-made measurement mode for the MycoAlert™ Mycoplasma Detection Assay for simple user interface, and can also be used for many other luminescence-based assays, such as cell proliferation and cytotoxicity assays or Luciferase reporter gene assays.

Easy-To-Use

- Can be used as a portable battery-operated instrument
- Lightweight instrument with small footprint
- No computer required

Two Basic Measurement Modes

- Tailor-made mode for running MycoAlert™ Mycoplasma Detection Assay
- Quick Single Read mode for unprocessed luminescence readings

High Sensitivity and Linearity

- Detection of less than 50 amol ATP or less than 1 000 luciferase molecules in one reporter gene assay
- Dynamic range of 5.5 orders of magnitude

High Data-Storage Capacity

- Internal memory with maximum storage of up to 1 500 single readings
- Optionally, sample results may be uploaded to a computer and viewed and analyzed with Microsoft® Excel®

Broad Range of Applications

- MycoAlert™ Mycoplasma Detection Assay
- ATP assays, e.g. ViaLight™ Cell Proliferation and Cytotoxicity Assay
- Adenylate Kinase assays, e.g. ToxiLight™ Non-destructive Cytotoxicity Assay
- Luciferase reporter gene analysis

Technical Specifications

Dimensions (w × d × h)	175 × 112 × 140 mm (6.89 × 4.41 × 5.51 in)
Weight	≈1.35 kg (2.98 lbs)
Power supply	Input: 100–240 V AC Output: 7.5 V DC/1.7 A



Figure 2
Lucetta™ Luminometer for sensitive luminescence measurements.

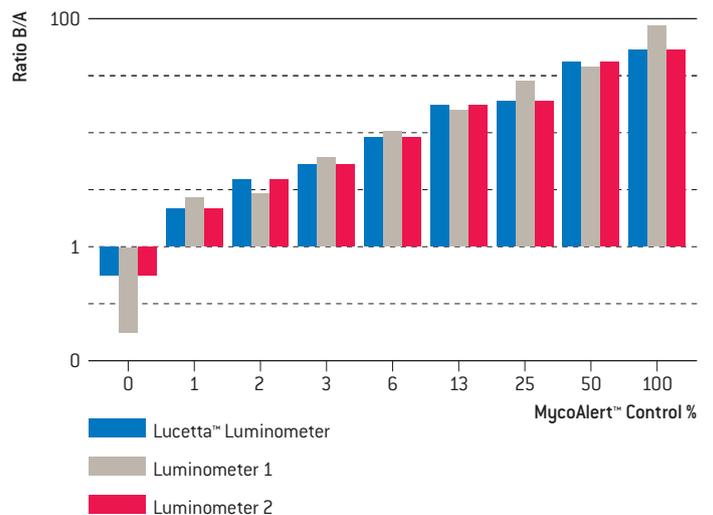


Figure 3
Dose response curve of the Lucetta™ Luminometer using MycoAlert™ Assay Control. A MycoAlert™ Mycoplasma Detection Assay was conducted for samples containing different amounts of positive MycoAlert™ Assay Control. Measurements were performed in parallel using three different single tube luminometers.

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

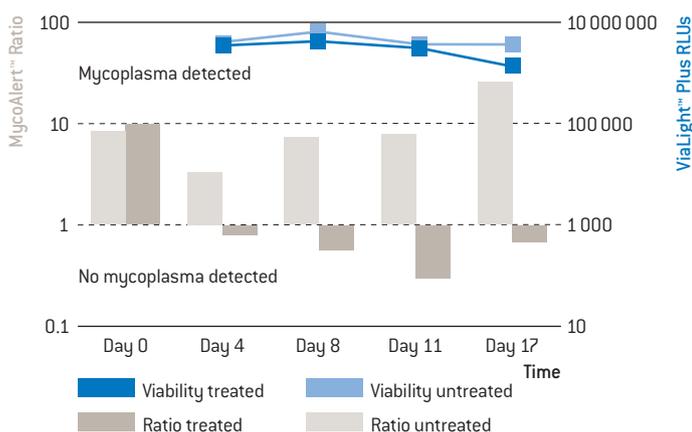


Figure 3
Effect of MycoZap™ Elimination Reagent on cell viability and mycoplasma removal. The MycoZap™ Reagent treatment eliminates mycoplasma in as few as 4 days with minimal impact on cell viability. Mycoplasma detection was performed with MycoAlert™ Assay and viability was determined by ViaLight™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

MycoZap™ Spray (Available in EU only)

The new MycoZap™ Spray is an effective solution for reliable disinfection of laboratory surfaces and apparatus, such as clean benches, incubators, cell storage boxes and liquid nitrogen containers. MycoZap™ Spray shows an excellent effectiveness against mycoplasma. A special biological ingredient shows an extreme affinity to mycoplasma and develops its full activity together with the alcoholic and detergent properties of MycoZap™ Spray. Clean and contamination-free surfaces can be achieved in a single application.

- Eradicates mycoplasma within seconds
- Non-corrosive and non-carcinogenic
- Effective against a broad range of organisms including enveloped and non-enveloped viruses, Gram (+) and Gram (-) bacteria as well as protozoa
- Convenient, ready-to-use and stable

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
Lucetta™ Luminometer		
AAL-1001	Lucetta™ Luminometer	—
AWL-1002	Lucetta™ Guarantee, 2-year extension	—
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
VZA-2001	MycoZap™ Spray*	500 ml
VZA-2002	MycoZap™ Spray (refill)*	1 000 ml

* MycoZap™ Spray is only available in Austria, Belgium, Denmark, France, Germany, Italy, Luxembourg, Netherlands, Poland, Spain, Sweden, Switzerland, UK and distributed countries on the European continent. For availability in other countries, please contact our Customer Service (for contact details please refer to back page).

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

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