

## **Mycoplasma free Cell Culture using Biontex products**

### **Frequently asked questions**

#### **Must I run a mycoplasma contamination test after each standard application of MycoRAZOR® (5 passes)?**

Yes. It is important to check that mycoplasmas have been completely eliminated (e.g. by using MycoSPY™) to prevent the establishment of resistance. As resistance can be built up in the same way as in all use of antibiotics, complete elimination of mycoplasmas is vital.

#### **After one application cycle (3 passes) of MycoRAZOR® a mycoplasma test showed positive. What action should I take?**

First check whether two passes without the use of MycoRAZOR® were conducted between the last use of MycoRAZOR® and the current test. If this was not the case, dead mycoplasma may have been detected, particularly by highly sensitive methods such as PCR. If the minimum time between the last application and the test was observed, use MycoRAZOR® in the next five passes of your cells, gradually increasing the dose of MycoRAZOR® up to a maximum dilution of 1/25. Please note that depending on the cell type, toxic effects may result from increasing the concentration of MycoRAZOR®. If you observe toxic effects (lower proliferation rate; changes in cell morphology) in your cells, apply the last-used dose in the remaining cycles. It is essential to test for results of each treatment by using a mycoplasma detection kit!

#### **After the successful use of MycoRAZOR® and testing free of mycoplasma, a culture has developed further mycoplasma contamination. How did this happen?**

The animal products used in cell culture are primary sources of mycoplasma contamination. To avoid this risk, use only fetal bovine serum (FBS) and trypsin that are guaranteed mycoplasma free.

Mycoplasmas belong to the class of Mollicutes and thus lack cell walls, they are resistant to many antibiotics that attack cell wall synthesis. The user is thus an important source of contamination in routine use of this type of antibiotic for cell culture. In this case, non-sterile working conditions go unnoticed, as the addition of antibiotics prevents the growth of most bacteria – and thus macroscopic effects – while allowing mycoplasmas to multiply unhindered.

In addition, cross-contamination from another cell culture is possible. For this reason always test all cultured cells and replace any potentially contaminated cell culture material (medium, FBS, trypsin, buffer, LN2 for storage).