

Y. pestis Bacterium qPCR Detection Kit (PLAG) Catalog #RU7088 100 reactions

Product Description

Yersinia pestis (*Y. pestis*) is a non-spore-forming, Gram-negative, non-motile, and rod-shaped bacterium. It can infect humans and other mammals via rat fleas and causes pneumonic, septicemic, and bubonic plague. Plague can be transmitted among humans and mammals by contacting with contaminated fluid or tissue or breathing in *Y. pestis*-containing droplets. ScienCell's *Y. pestis* Bacterium qPCR Detection Kit (PLAG) is designed to detect the presence of *Y. pestis* bacterium in extracted DNA samples. The plague primer set (Cat #7088-PLAG) included in the kit targets *Y. pestis* genome. It is verified to have high specificity and efficiency near 100% under recommended PCR conditions. In addition, a non-infectious positive control (Cat #7088-Pos) and nuclease-free water (Cat #GQ100-4) are included in the kit. The positive control consists of non-infectious DNA fragments and serves to ensure reagents and instruments are working properly. Please refer to Tables 4 and 5 for results interpretation.

Kit Components

Cat #	Component	Quantity	Storage
MB6018a-1	2X GoldNStart TaqGreen qPCR master mix	1 mL	-20°C
7088-PLAG	Plague primer set, in solution 200 μL -2		-20°C
7088-Pos	Positive control (non-infectious DNA: 1,000-2,000 copies/µL)	200 µL	-80°C
GQ100-4	Nuclease-free H ₂ O	4 mL	4°C

Additional Materials Required (Materials Not Included in Kit)

Component	Recommended
DNA extraction kit	SpeeDNA Isolation Kit (ScienCell, Cat #MB6918)
qPCR plate or tube	

Quality Control

The primer set and the positive control are validated by qPCR using serially diluted templates. The PCR products are analyzed by gel electrophoresis.

Product Use

For Research Use Only. Not for use in diagnostic procedures.

Shipping and Storage

The product is shipped on dry ice. Upon receipt, store the 2X GoldNStart TaqGreen qPCR master mix (Cat #MB6018a-1), the primer set (Cat #7088-PLAG), and the positive control (Cat #7088-Pos) at -20°C in a manual defrost freezer, and nuclease-free H₂O (Cat #GQ100-4) at 4°C. Aliquot as needed. Avoid repeated freeze-and-thaw cycles.

Procedures

Important: Only use nuclease-free reagents in PCR applications.

Note: 2X GoldNStart TaqGreen qPCR master mix (Cat #MB6018a-1) does not contain a ROX passive reference dye. If the qPCR instrument being used has a "ROX passive reference dye" option, please deselect this option.

- 1. Prior to use, allow the primer set (Cat #7088-PLAG) to thaw to room temperature. Shake gently to mix well.
- 2. Centrifuge the vials at 1,500x g for 1 minute.
- 3. Aliquot the primer set as needed. Store at -20°C in a manual defrost freezer. Avoid repeated freeze-and-thaw cycles. Keep on ice when thawed.
- 4. With the test samples, two control samples should be run concurrently: the non-infectious positive control (Cat #7088-Pos) and H₂O (Cat #GQ100-4) as the No Template Control (NTC). Prepare one qPCR reactions for each control sample with #7088-PLAG primer set. Prepare 20 μl qPCR reactions as shown in Table 1.

Table 1.

Total volume	20 µl
Nuclease-free H ₂ O (Cat #GQ100-4)	3 µl
2X GoldNStart TaqGreen qPCR master mix (Cat # MB6018a-1)	
Primer set (Cat #7088-PLAG)	2 µl
Control sample (Cat #7088-Pos or GQ100-4)	5 µl

5. For each extracted DNA test sample, prepare one qPCR reaction with #7088-PLAG primer set. Prepare 20 µl qPCR reactions as shown in Table 2.

Table 2.

DNA test sample (concentration varies)	5 µl
Primer set (Cat #7088-PLAG)	2 µl
2X GoldNStart TaqGreen qPCR master mix (Cat # MB6018a-1)	
Nuclease-free H ₂ O (Cat #GQ100-4)	3 µl
Total volume	20 µl

- 6. Seal the qPCR reaction wells. Centrifuge the plates or tubes at 1,500x g for 15 seconds.
- 7. Setup qPCR reactions as shown in Table 3.

Table 3. Instrument settings for qPCR reactions.

Fluorescence data should be collected during the data acquisition step.

Step	Temperature	Time	Number of cycles
Taq DNA polymerase activation	95°C	10 min	1

Denaturation	95°C	20 sec	
Annealing	65°C	20 sec	40
Extension	72°C	20 sec	40
Data acquisition	Pl	ate read	

Results Interpretation

Table 4. Interpretation of PLAG kit control sample test results.

Sample	7088-PLAG	Results Interpretation
7088 Dec	+	Expected
7088-Pos	-	qPCR failed
CO100 4	-	Expected
GQ100-4	+	Reagent(s) contaminated

Note: Cq values less than 35 are considered positive. Cq values equal to or greater than 35 are considered negative.

Table 5. Interpretation of PLAG kit target sample test results when control results are as expected.

7088-PLAG	Results Interpretation
+	Y. pestis bacterium detected
-	No Y. pestis bacterium detected

Note: Cq values less than 35 are considered positive. Cq values equal to or greater than 35 are considered negative.