

TrueMark™ Pre-Designed and Custom Multiplex Panels

USER GUIDE

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For descriptions of symbols on product labels or product documents, go to [thermofisher.com/symbols-definition](https://www.thermofisher.com/symbols-definition).

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Revision	Date	Description
A	19 August 2024	New <i>TrueMark™ Pre-Designed and Custom Multiplex Panels User Guide</i> .

The information in this guide is subject to change without notice.

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Product information

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IMPORTANT! Before using this product, read and understand the information in the “Safety” appendix in this document.

Product description

TrueMark™ Multiplex Panels are a collection of real-time PCR assays that use spectral multiplexing to detect microbes and internal controls simultaneously in the same reaction. Panels in the following disease areas are available in 200- and 1,000-reaction sizes, and can be ordered in a single-tube tube format or as part of TrueMark™ Custom Plates.

- Antibiotic resistance
- Gastrointestinal infection
- Bloodborne pathogen
- Meningitis
- Nail fungus
- Oncology
- Respiratory tract microbiota
- Sexually transmitted infection
- Soft tissue infection
- Transplant
- Urinary tract infection
- Vaginal tract microbiota
- Vector-borne

TrueMark™ Multiplex Panels include pre-designed content or can be customized to meet individual laboratory needs.

For more information about the panels, or to request a quote, contact your sales representative, or visit thermofisher.com/customtruemark.

For information on TrueMark™ Custom Plates, see “Related documentation” on page 25.

TrueMark™ Pre-Designed Multiplex Panels

- Pre-designed panels are in-silico tested for inclusivity and interaction with high-risk oligos. A subset of pre-designed panels also undergo functional testing, including analytical sensitivity, linear dynamic range, and competitive interference (if applicable).
- For a list of available and qualified TrueMark™ Pre-Designed Multiplex Panels, refer to [thermofisher.com/truemarkpredesigned](https://www.thermofisher.com/truemarkpredesigned).
- For contents and storage information for each TrueMark™ Pre-Designed Multiplex Panel, see the panel product information sheet.

TrueMark™ Custom Multiplex Panels

- For a list of available species and assays, use the assay search tool: [thermofisher.com/microbe-detection/taqman/query/](https://www.thermofisher.com/microbe-detection/taqman/query/).

Required materials not supplied

Unless otherwise indicated, all materials are available through [thermofisher.com](https://www.thermofisher.com). "MLS" indicates that the material is available from [fisherscientific.com](https://www.fisherscientific.com) or another major laboratory supplier.

Catalog numbers that appear as links open the web pages for those products.

Item	Source
Real-time PCR instrument, one of the following or equivalent	
Applied Biosystems™ QuantStudio™ 5 Real-Time PCR System (96-well, 0.2 mL block)	A28569 (with laptop) A28574 (with desktop) A28139 (standalone)
Applied Biosystems™ QuantStudio™ 7 Flex Real-Time PCR System (384-well)	4485695 (with laptop) 4485701 (with desktop)
Recommended software	
Design and Analysis Software v2.7 or later	thermofisher.com/qpcrsoftware
Equipment	
BSL-2 biological safety cabinet	MLS
Centrifuge, with a rotor that can accommodate standard microplates	MLS
Laboratory freezers (-30°C to -10°C)	MLS
Laboratory mixer, vortex or equivalent	MLS
Microcentrifuge	MLS
Refrigerator (2°C to 8°C)	MLS
Single-channel and multichannel adjustable pipettors (2 µL to 1,000 µL)	MLS

(continued)

Item	Source
Master mix, one of the following	
TaqPath™ 1-Step Multiplex Master Mix (No ROX™)	A28521 , A28522 , A28523
TaqPath™ DuraPlex™ 1-Step RT-qPCR Master Mix (No ROX™)	A58669 , A58670 , A58671
Amplification control, one of the following	
TrueMark™ Custom DNA Control	A50319 , A50375 , A50376 , A50377 , A50378 , A50379 , A50380 , A50381
TrueMark™ Amplification Control I (5 × 10 ⁷ copies/μL)	A55698
TrueMark™ Amplification Control I (1 × 10 ⁵ copies/μL)	A55699
TrueMark™ Comprehensive Microbiota Control (1 × 10 ⁵ copies/μL)	A50382
TrueMark™ Comprehensive Microbiota Control (5 × 10 ⁷ copies/μL)	A50383
Reagents	
Nuclease-free water (not DEPC-treated)	AM9932 , AM9938 , or MLS
TE Buffer (10 mM Tris-HCl [pH 8.0], 0.1 mM EDTA)	12090015 or MLS
TrueMark™ Universal Extraction Control Organism (<i>B. atrophaeus</i>)	A39180
Consumables	
MicroAmp™ Optical 96-Well Reaction Plate	N8010560 4316813
MicroAmp™ Optical 96-Well Reaction Plate with Barcode	4306737 , 4326659
MicroAmp™ Optical 384-Well Reaction Plate	4343370
MicroAmp™ Optical 384-Well Reaction Plate with Barcode	4309849 , 4326270 , 4343814
MicroAmp™ Fast Optical 96-Well Reaction Plate, 0.1 mL	4346907
MicroAmp™ Fast Optical 96-Well Reaction Plate with Barcode, 0.1 mL	4346906 , 4366932
MicroAmp™ Optical Adhesive Film	4311971 , 4360954
MicroAmp™ Optical Film Compression Pad ^[1]	4312639
Microcentrifuge tubes, non-stick, RNase-free (1.5-mL and 2-mL)	thermofisher.com/plastics
Pipette tips, sterile, filtered (aerosol barrier)	thermofisher.com/pipettetips
Reagent reservoirs for multichannel pipettes	MLS

^[1] Required for use with the QuantStudio™ 5 Real-Time PCR Instrument (96-well, 0.2 mL block).

Samples and controls

- Samples should always be treated as if infectious and/or biohazardous in accordance with safe laboratory procedures.
- Samples must be collected, transported, and stored according to appropriate laboratory guidelines.
- Positive and negative controls must be included to accurately interpret assay results.

Table 1 Controls

Control	Description	Recommended sample type
Ribonuclease P (RNase P)	Endogenous sample collection control for the respiratory tract infection (RTI), sexually transmitted infection (STI), lesion, vaginal, and genital infection panels; monitors sample quality and nucleic acid extraction.	Swabs
<i>Bacillus atrophaeus</i> (TrueMark™ Universal Extraction Control Organism (<i>B. atrophaeus</i>))	Exogenous process control for the urinary tract infection (UTI) panel and the gastrointestinal infection (GI) panel that is added to each sample during nucleic acid extraction; monitors nucleic acid extraction and absence of real-time PCR inhibitors.	Urine, stool
Amplification control ^[1]	A positive control on the real-time PCR plate that monitors real-time PCR reaction setup and reagent integrity.	—
No template control (NTC)	A negative control on the real-time PCR plate that contains Reaction Mix and water only; monitors contamination during real-time PCR reaction setup.	—
(Recommended) Negative extraction control (NEC)	A negative control on the nucleic acid extraction plate that contains water or buffer instead of a sample. The extracted NEC is run on the real-time PCR plate to monitor cross-contamination during the nucleic acid extraction and the absence of real-time PCR inhibitors.	—

^[1] See “Required materials not supplied” on page 6.

In-use reagent stability

Reagent	Stability information
TrueMark™ Custom Multiplex Panel	Once thawed, the panels are stable for up to 8 hours at 2°C to 8°C. Do not exceed 5 freeze-thaw cycles.
Assembled real-time PCR reaction plate	For best results, we recommend that you run the assembled real-time PCR reaction plate immediately. The assembled real-time PCR reaction plate is stable for up to 60 minutes at 2–8°C.



Before you begin

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- General laboratory guidelines 9

Recommended real-time PCR instruments and software

The following real-time PCR instruments are recommended for each plate format. Design and Analysis Software v2.7 or later is recommended for instrument setup and data analysis. It is the responsibility of the laboratories to validate their own real-time PCR instrument parameters for each panel.

Table 2 Recommended instruments and software

Plate format	Instrument	Software
96-well	QuantStudio™ 5 Real-Time PCR System (page 16)	Design and Analysis Software v2.7 or later
384-well	QuantStudio™ 7 Flex Real-Time PCR System (page 18)	

Dye calibration

Ensure that your real-time PCR instrument is calibrated for the dyes used in your assay and that the instrument calibrations are current. For more information, refer to the instrument user guide (see “Related documentation” on page 25).

General laboratory guidelines

- Implement standard operating procedures in your laboratory to prevent contamination, such as the following:
 - Frequent glove changes
 - Frequent decontamination of surfaces, equipment, and pipettes with fresh 10% bleach (1% V/V sodium hypochlorite) or decontamination solution, followed by 70% ethanol
 - Use of ultraviolet light during biosafety cabinet decontamination (when available)
- To prevent degradation, keep samples, master mixes, assays, and controls on ice or in cold blocks while in use.
- To support reliable reagent performance, do not exceed 5 freeze-thaw cycles.

- To prevent stock contamination and reduce the number of freeze-thaw cycles, aliquot reagents using low DNA-binding tubes.
- After each run, review the amplification curves in the instrument software according to data QC standard operating procedures for your lab.
- To support reliable performance of the real-time PCR instrument, perform preventive maintenance according to the instructions provided by the instrument manufacturer (see “Related documentation” on page 25).

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Prepare real-time PCR reactions

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Guidelines for real-time PCR

IMPORTANT!

- Prepare and keep the real-time PCR plate on ice or in a cold block until it is loaded into the real-time PCR instrument.
 - For best results, run the real-time plate immediately after preparation. If the assembled reaction plate is not loaded into the real-time PCR instrument immediately, the plate is stable up to 1 hour at 4°C. Improper storage of the real-time PCR plate can result in degradation of samples.
 - To prevent contamination, prepare reagents in a PCR workstation or equivalent amplicon-free area. Do not use the same pipette for controls and samples, and always use aerosol barrier pipette tips.
 - Maintain a nuclease-free environment. Periodically decontaminate surfaces.
 - Protect assays from light.
 - Keep samples and components on ice or in a cold block during use.
 - For each real-time PCR plate, include the following controls:
 - One amplification control (see “Required materials not supplied” on page 6)
 - One no template control (NTC)
 - One negative extraction control (NEC)
-

Prepare real-time PCR reactions: 96-well reaction plate

Before you begin:

- If frozen, thaw the samples and reagents on ice or in a cold block.
 - If freshly extracted, keep the samples on ice or in a cold block.
1. Gently vortex the reagents, then briefly centrifuge the tube or swirl the bottle to collect the liquid at the bottom of the container. Keep the reagents on ice until use.
 2. Dilute the amplification control to the working concentration. Keep the amplification control on ice until use.

Note: The optimal amplification control concentration can vary and should be determined for each assay by the laboratory.

3. Prepare the reaction mix by combining the following components sufficient for the number of samples, at least one amplification control, one NTC, one NEC, and the overage.

Note: If improved sensitivity is needed, a total of 14 μL of the sample elute volume can be added to the real-time PCR reaction by replacing 9 μL of water from the reaction mix with an additional 9 μL of the sample elute (see substep 4b).

Component	Volume per sample or control	Volume for n samples plus controls ^[1,2]
Master mix (see “Required materials not supplied” on page 6)	5 μL	$5 \times (1.2 \times n)$ μL
TrueMark™ Custom Multiplex Panel (20X)	1 μL	$1 \times (1.2 \times n)$ μL
Nuclease-free water (not DEPC-treated)	9 μL	$9 \times (1.2 \times n)$ μL
Total reaction mix volume	15 μL	—

^[1] All volumes include 20% overage for pipetting error.

^[2] 'n' includes samples, NTC, and NEC (if used).

4. Set up the real-time PCR reaction plate.

Note: Prepare the plate on ice or a cold block.

- Pipet 15 μL of the reaction mix prepared in step 3 to each well of a MicroAmp™ Optical 96-Well Reaction Plate.
- Add 5 μL of either the extracted samples, the diluted amplification control, nuclease-free water (NTC), or NEC to the designated wells.

Note: Up to 14 μL of sample eluate can be added. For more information, see step 3.

- Seal the plate thoroughly with MicroAmp™ Optical Adhesive Film.

IMPORTANT!

- Use ONLY MicroAmp™ Optical Adhesive Film (Cat. No. [4311971](#), [4360954](#)).
- DO NOT use optical caps, MicroAmp™ Clear Adhesive Film, or any other film or sealing method.
- DO NOT heat seal the plate.
- When applying the MicroAmp™ Optical Adhesive Film, ensure that pressure is applied across the entire plate and that there is a tight seal across every individual well. Failure to do so runs the risk of an improperly sealed well, leading to potential well-to-well contamination during vortexing and evaporation during PCR.

5. Vortex the plate on a flat rubber surface at the highest setting speed for 30–60 seconds as follows: 5–10 seconds in the center of the plate, 5–10 seconds in each plate corner, and 5–10 seconds again in the center.

IMPORTANT! Move the plate around to ensure equal contact of each part of the plate with the vortex mixer platform.

6. Centrifuge the reaction plate for 1-2 minutes at $\geq 650 \times g$ to remove bubbles and to collect the liquid at the bottom of the reaction plate.

For best results, proceed immediately to real-time PCR (Chapter 4, “Set up and run the real-time PCR instrument”).

IMPORTANT!

- Keep the real-time PCR reaction plate at 2°C to 8°C until it is loaded into the real-time PCR instrument immediately before starting the run.
 - Run the real-time PCR reaction plate within 1 hour after preparation. Failure to do so could result in degradation of samples.
-

Prepare real-time PCR reactions: 384-well reaction plate

Before you begin:

- If frozen, thaw the samples and reagents on ice or in a cold block.
 - If freshly extracted, keep the samples on ice or in a cold block.
1. Gently vortex the reagents, then briefly centrifuge the tube or swirl the bottle to collect the liquid at the bottom of the container. Keep the reagents on ice until use.
 2. Dilute the amplification control to the working concentration. Keep the amplification control on ice until use.

Note: The optimal amplification control concentration can vary and should be determined for each assay by the laboratory.

3. Prepare the reaction mix by combining the following components sufficient for the number of samples, at least one amplification control, one NTC, one NEC, and the overage.

Note: If improved sensitivity is needed, a total of 7 μL of the sample elute volume can be added to the real-time PCR reaction by replacing 2 μL of water from the reaction mix with an additional 2 μL of the sample elute (see substep 4b).

Component	Volume per sample or control	Volume for n samples plus controls ^[1,2]
Master mix (see “Required materials not supplied” on page 6)	2.5 μL	$2.5 \times (1.2 \times n)$ μL
TrueMark™ Custom Multiplex Panel (20X)	0.5 μL	$0.5 \times (1.2 \times n)$ μL
Nuclease-free water (not DEPC-treated)	2 μL	$2 \times (1.2 \times n)$ μL
Total reaction mix volume	5 μL	—

^[1] All volumes include 20% overage for pipetting error.

^[2] 'n' includes samples, NTC, and NEC (if used).

4. Set up the real-time PCR reaction plate.

Note: Prepare the plate on ice or a cold block.

- Pipet 5 μL of the reaction mix prepared in step 3 to each well of a MicroAmp™ Optical 384-Well Reaction Plate.
- Add 5 μL of either the extracted samples, the diluted amplification control, nuclease-free water (NTC), or NEC to the designated wells.

Note: Up to 7 μL of sample eluate can be added. For more information, see step 3.

- Seal the plate thoroughly with MicroAmp™ Optical Adhesive Film.

IMPORTANT!

- Use ONLY MicroAmp™ Optical Adhesive Film (Cat. No. [4311971](#), [4360954](#)).
- DO NOT use optical caps, MicroAmp™ Clear Adhesive Film, or any other film or sealing method.
- DO NOT heat seal the plate.
- When applying the MicroAmp™ Optical Adhesive Film, ensure that pressure is applied across the entire plate and that there is a tight seal across every individual well. Failure to do so runs the risk of an improperly sealed well, leading to potential well-to-well contamination during vortexing and evaporation during PCR.

5. Vortex the plate on a flat rubber surface at the highest setting speed for 30–60 seconds as follows: 5–10 seconds in the center of the plate, 5–10 seconds in each plate corner, and 5–10 seconds again in the center.

IMPORTANT! Move the plate around to ensure equal contact of each part of the plate with the vortex mixer platform.

6. Centrifuge the reaction plate for 1-2 minutes at $\geq 650 \times g$ to remove bubbles and to collect the liquid at the bottom of the reaction plate.

For best results, proceed immediately to real-time PCR (Chapter 4, “Set up and run the real-time PCR instrument”).

IMPORTANT!

- Keep the real-time PCR reaction plate at 2°C to 8°C until it is loaded into the real-time PCR instrument immediately before starting the run.
 - Run the real-time PCR reaction plate within 1 hour after preparation. Failure to do so could result in degradation of samples.
-

4

Set up and run the real-time PCR instrument

- Set up and run the QuantStudio™ 5 Real-Time PCR System (96-well, 0.2 mL block) 16
- Set up and run the QuantStudio™ 7 Flex Real-Time PCR System (384-well block) 18

For instrument and software recommendations, see Chapter 2, “Before you begin”.

Set up and run the QuantStudio™ 5 Real-Time PCR System (96-well, 0.2 mL block)

1. From the main screen of the Design and Analysis Software v2.7 or later, select **Set Up Plate**.
2. In the **Systems Templates** tab, select the **Quantification-Taqman** template with the following properties.
 - **Instrument: QuantStudio™ 5**
 - **Block: 96-Well 0.2-mL**
 - **Run Mode: Standard**
 - **Analysis: Standard Curve**
3. In the **Run Method** tab, set up the thermal protocol for your instrument.
 - **Reaction Volume: 20 µL**
 - **Heated Cover Temperature: 105.0°C**

Table 3 QuantStudio™ 5 Real-Time PCR Instrument, 96-well, 0.2-mL block

Step	Temperature	Time	Number of cycles	Ramp rate
UNG incubation	25°C	2 minutes	1	1.6°C per second
Reverse transcription ^[1]	53°C	10 minutes	1	1.6°C per second
Preincubation	85°C	10 minutes	1	1.6°C per second
Activation	95°C	2 minutes	1	1.6°C per second
Denaturation	95°C	3 seconds	40	1.6°C per second
Annealing / extension	60°C	32 seconds		1.6°C per second

^[1] Required for panels with RNA targets only.

4. Set up the optical filters for your instrument.

Table 4 Default PCR filters for the QuantStudio™ 5 Real-Time PCR Instrument, 96-well, 0.2-mL block

		Emission Filter					
		m1(520±15)	m2(558±11)	m3(586±10)	m4(623±14)	m5(682±14)	m6(711±12)
Excitation Filter	x1(470±15)	✓	✓	✓			
	x2(520±10)		✓	✓	✓		
	x3(550±11)			✓	✓		
	x4(580±10)				✓	✓	
	x5(640±10)					✓	
	x6(662±10)						

5. In the **Plate Setup** tab, confirm that the **Passive Reference** is set to **None**.

6. In the **Targets** table, confirm the targets, reporter dyes, probe types, and quencher selections.

Note: Depending on the panel, the list of targets and their corresponding reporter dyes will vary. Only add the targets and dyes that are present in the panel.

Target	Reporter dye	Quencher
Target A	FAM™	None
Target B	VIC™	
Target C	ABY™	
Target D	JUN™	
Internal control	Cy5™	

IMPORTANT! Change the target names according to the panel tested.

7. Confirm that the targets in step 6 are assigned to each well in the plate layout.

8. Save the plate file as EDT, then load the file onto the instrument.

- Place a MicroAmp™ Optical Film Compression Pad with the gray side down on the surface of the real-time PCR reaction plate, ensuring a proper seal between the thermal cycler and the adhesive film.

IMPORTANT!

- Be careful to place the compression pad with the brown side up and the gray side down, centered on top of the plate.
 - Ensure the compression pad is free from wrinkles and signs of deterioration prior to use.
 - Each compression pad can be used up to 20 times before discarding. Do not use more than 20 times. Use of deteriorated compression pads can lead to inconsistencies in reaction volumes and reaction failure.
-

- Load the plate onto the instrument, then touch **Start Run**. When prompted, confirm that you inserted the plate.

Set up and run the QuantStudio™ 7 Flex Real-Time PCR System (384-well block)

- From the main screen of the Design and Analysis Software v2.7 or later, select **Set Up Plate**.
- In the **Systems Templates** tab, select the **Quantification-Taqman** template with the following properties.
 - Instrument:** QuantStudio™ 7 Flex
 - Block:** 384-Well
 - Run Mode:** Standard
 - Analysis:** Standard Curve
- In the **Run Method** tab, set up the thermal protocol for your instrument.
 - Reaction Volume:** 10 µL
 - Heated Cover Temperature:** 105.0°C

Table 5 QuantStudio™ 7 Flex Real-Time PCR Instrument, 384-well block

Step	Temperature	Time	Number of cycles	Ramp rate
UNG incubation	25°C	2 minutes	1	1.6°C per second
Reverse transcription ^[1]	53°C	10 minutes	1	1.6°C per second
Preincubation	85°C	10 minutes	1	1.6°C per second
Activation	95°C	2 minutes	1	1.6°C per second
Denaturation	95°C	3 seconds	40	1.6°C per second
Annealing / extension	60°C	39 seconds		1.6°C per second

^[1] Required for panels with RNA targets only.

4. Set up the optical filters for your instrument.

Table 6 Default PCR filters for the QuantStudio™ 7 Flex Real-Time PCR Instrument, 384-well block

		Emission Filter					
		m1(520±15)	m2(558±11)	m3(586±10)	m4(623±14)	m5(682±14)	m6(711±12)
Excitation Filter	x1(470±15)	✓	✓	✓			
	x2(520±10)		✓	✓	✓		
	x3(550±11)			✓	✓		
	x4(580±10)				✓	✓	
	x5(640±10)					✓	
	x6(662±10)						

5. In the **Plate Setup** tab, confirm that the **Passive Reference** is set to **None**.

6. In the **Targets** table, confirm the targets, reporter dyes, probe types, and quencher selections.

Note: Depending on the panel, the list of targets and their corresponding reporter dyes will vary. Only add the targets and dyes that are present in the panel.

Target	Reporter dye	Quencher
Target A	FAM™	None
Target B	VIC™	
Target C	ABY™	
Target D	JUN™	
Internal control	Cy5™	

IMPORTANT! Change the target names according to the panel tested.

7. Confirm that the targets in step 6 are assigned to each well in the plate layout.
8. Save the plate file as EDT, then load the file onto the instrument.
9. Load the plate onto the instrument. Ensure there is a proper seal between the thermal cycler and the adhesive film.
10. Touch **Start Run**. When prompted, confirm that you inserted the plate.



Analyze data

IMPORTANT! It is the responsibility of the laboratories using the TrueMark™ Custom Multiplex Panels to design and validate their own experiment and analysis parameters.

(Recommended) Use Design and Analysis Software v2.7 or later for data analysis. For more information about using the software, see “Related documentation” on page 25.

Note: For detailed information about data analysis, see the appropriate documentation for your instrument.

1. In the Design and Analysis Software v2.7 or later home screen, open the data file (EDS).
 2. In the open data file, click **Actions** ▶ **Save As**, then save the data file with a unique name.
 3. In the analysis settings, select automatic baseline with a start cycle of **5** and an end cycle of **auto**, or select according to lab validation.
 4. Set the appropriate threshold values for each target, as validated by your laboratory.
-

IMPORTANT! Do not use automatic threshold values.

5. Determine C_q cutoff values for each target for samples and controls.
-

Note: Design and Analysis Software v2.7 or later reports C_q values instead of C_t values. The C_q values are equivalent to C_t values.

6. Analyze results according to analysis, interpretation, and QC parameters, as validated by your laboratory.

For more information, contact support (“Customer and technical support” on page 25).



Supplemental information

TrueMark™ Universal Extraction Control Organism (*B. atrophaeus*)

TrueMark™ Universal Extraction Control Organism (*B. atrophaeus*) (Cat. No. [A39180](#)), contains three vials of lyophilized *Bacillus atrophaeus*, for use as a process control for cell lysis and nucleic acid recovery. This control is used with the proprietary TaqMan™ Assay for *Bacillus atrophaeus* sequences.

Like other gram-positive bacteria, *Bacillus atrophaeus* has thick cell walls that can be difficult to lyse. This characteristic makes *Bacillus atrophaeus* an ideal control to monitor the efficiency of cell lysis and subsequent nucleic acid recovery.

TrueMark™ Universal Extraction Control Organism (*B. atrophaeus*) is supplied lyophilized with a quantity of 1×10^9 copies/vial, and is reconstituted in 200 μL of 1X PBS (1X), pH 7.4 to a final concentration 5×10^6 copies/ μL .

The following *B. atrophaeus* dilution and the spike in process instructions are recommended:

Vortex for 30 seconds, until the pellet is resuspended. Make the suitable number of aliquots and stored at -20°C . Perform further dilution to obtain the working concentration of 5×10^4 copies/ μL .

10 μL of the control is added to urine samples in each extraction well at the start of extraction.



Safety

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WARNING! GENERAL SAFETY. Using this product in a manner not specified in the user documentation may result in personal injury or damage to the instrument or device. Ensure that anyone using this product has received instructions in general safety practices for laboratories and the safety information provided in this document.

- Before using an instrument or device, read and understand the safety information provided in the user documentation provided by the manufacturer of the instrument or device.
- Before handling chemicals, read and understand all applicable Safety Data Sheets (SDSs) and use appropriate personal protective equipment (gloves, gowns, eye protection, and so on). To obtain SDSs, visit [thermofisher.com/support](https://www.thermofisher.com/support).

Chemical safety



WARNING! GENERAL CHEMICAL HANDLING. To minimize hazards, ensure laboratory personnel read and practice the general safety guidelines for chemical usage, storage, and waste provided below. Consult the relevant SDS for specific precautions and instructions:

- Read and understand the Safety Data Sheets (SDSs) provided by the chemical manufacturer before you store, handle, or work with any chemicals or hazardous materials. To obtain SDSs, see the "Documentation and Support" section in this document.
- Minimize contact with chemicals. Wear appropriate personal protective equipment when handling chemicals (for example, safety glasses, gloves, or protective clothing).
- Minimize the inhalation of chemicals. Do not leave chemical containers open. Use only with sufficient ventilation (for example, fume hood).
- Check regularly for chemical leaks or spills. If a leak or spill occurs, follow the manufacturer cleanup procedures as recommended in the SDS.
- Handle chemical wastes in a fume hood.
- Ensure use of primary and secondary waste containers. (A primary waste container holds the immediate waste. A secondary container contains spills or leaks from the primary container. Both containers must be compatible with the waste material and meet federal, state, and local requirements for container storage.)
- After emptying a waste container, seal it with the cap provided.
- Characterize (by analysis if needed) the waste generated by the particular applications, reagents, and substrates used in your laboratory.
- Ensure that the waste is stored, transferred, transported, and disposed of according to all local, state/provincial, and/or national regulations.
- **IMPORTANT!** Radioactive or biohazardous materials may require special handling, and disposal limitations may apply.



WARNING! HAZARDOUS WASTE (from instruments). Waste produced by the instrument is potentially hazardous. Follow the guidelines noted in the preceding General Chemical Handling warning.



WARNING! 4L Reagent and Waste Bottle Safety. Four-liter reagent and waste bottles can crack and leak. Each 4-liter bottle should be secured in a low-density polyethylene safety container with the cover fastened and the handles locked in the upright position.

Biological hazard safety



WARNING! Potential Biohazard. Depending on the samples used on this instrument, the surface may be considered a biohazard. Use appropriate decontamination methods when working with biohazards.



WARNING! BIOHAZARD. Biological samples such as tissues, body fluids, infectious agents, and blood of humans and other animals have the potential to transmit infectious diseases. Conduct all work in properly equipped facilities with the appropriate safety equipment (for example, physical containment devices). Safety equipment can also include items for personal protection, such as gloves, coats, gowns, shoe covers, boots, respirators, face shields, safety glasses, or goggles. Individuals should be trained according to applicable regulatory and company/ institution requirements before working with potentially biohazardous materials. Follow all applicable local, state/provincial, and/or national regulations. The following references provide general guidelines when handling biological samples in laboratory environment.

- U.S. Department of Health and Human Services, *Biosafety in Microbiological and Biomedical Laboratories (BMBL)*, 6th Edition, HHS Publication No. (CDC) 300859, Revised June 2020
www.cdc.gov/labs/pdf/CDC-BiosafetymicrobiologicalBiomedicalLaboratories-2020-P.pdf
- Laboratory biosafety manual, fourth edition. Geneva: World Health Organization; 2020 (Laboratory biosafety manual, fourth edition and associated monographs)
www.who.int/publications/i/item/9789240011311



Documentation and support

- Related documentation 25
- Customer and technical support 25

Related documentation

Document	Publication Number
<i>QuantStudio™ 3 and 5 Real-Time PCR Systems Installation, Use, and Maintenance Guide</i>	MAN0010407
<i>QuantStudio™ 6 and 7 Flex Real-Time PCR Systems Quick Reference</i>	4489826
<i>QuantStudio™ Design and Analysis Software 2 User Guide</i>	MAN0018200
<i>QuantStudio™ Design and Analysis Software 2 Standard Curve Analysis Module User Guide</i>	MAN0018746
<i>TaqMan™ Assay Multiplex PCR Optimization Application Guide</i>	MAN0010189
<i>TaqPath™ 1-Step Multiplex Master Mix User Guide</i>	MAN0014269
<i>TaqPath™ 1-Step Multiplex Master Mix Quick Reference</i>	MAN0014389
<i>TaqPath™ 1-Step Multiplex Master Mix Product Information Sheet</i>	MAN0014069, 100033995
<i>TrueMark™ Custom DNA Controls Product Information Sheet</i>	MAN0024963
<i>TrueMark™ Custom Plates User Guide</i>	MAN0025767
<i>TrueMark™ Universal Extraction Control Organism (B. atrophaeus) Product Information Sheet</i>	MAN0018535

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- Product documentation
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 - Certificates of Analysis
 - Safety Data Sheets (SDSs; also known as MSDSs)

Note: For SDSs for reagents and chemicals from other manufacturers, contact the manufacturer.
