# **Biological Water Testing**Reliable safety and quality control





## Ensure Safety and Quality With Reliable and Fast Biological Water Testing

Water is the basis of life. Drinking water, processing, and industrial water are relevant resources that need to be controlled and protected carefully.

Comprehensive water analysis is of major importance. Parameters such as total organic carbon (TOC), total nitrogen bound (TN $_{\rm b}$ ), and the analysis methods for mercury, trace, and toxic elements play an essential role. By supplying high performance TOC/TN $_{\rm b}$  analyzers as well as highly sensitive instruments, based on ICP-MS and ICP-OES, Analytik Jena offers a comprehensive solution for chemical water analysis. Protection from infections and unintentional contact with pathogens are also a vital part of water analysis. Reliable and efficient detection methods are essential for optimal action planning and management of hygiene and disinfection.

#### Your benefits

- Efficient from sample to result in a few hours instead of weeks
- Save reaction time results are available earlier
- Reliable specific detection kits secure results
- Optimized perfect combination of device and kit
- Comprehensive assays for numerous relevant microbiological targets

A conventional cultivating method takes up to 14 days, but only a few hours with Analytik Jena's PCR-based detection method for the highly specific and reliable microbiological parameters (see scheme 1).



#### General control parameter: overall bacterial load

The total bacterial load is a key indicator for the hygienic quality of water. It can indicate potential fouling of both, drinking and industrial water.\*

The innuDETECT Bacteria Quantification Assay is a semiquantitative assay which detects universal bacterial target genes simultanously with bacterial standard DNA also included in the assay. Internal Controls, co-amplified during the PCR, give maximum assurance on results and validate negative findings.

To demonstrate the excellent quality of the innuDETECT Bacterial Quantification Assay, it was used to analyze the general bacterial load in DNA extracts obtained from a 1 mL water sample. The results are shown in Figure 1. Table 1 demonstrates bacterial loads obtained from real-time PCR, based on exact ct values. Detected bacterial targets in an unknown sample can be quantified upon correlation with standard curves.



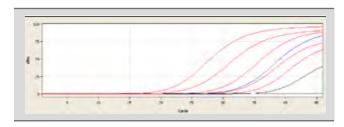


Figure 1/Table 1: Co-amplification of bacterial standard DNA (red) allows for semi-quantification of an unknown sample (blue) via ct values. Negative control is shown in black.

Sample	Ct value	General bacterial load/reaction
Standard 1	21.5	1 x 10 <sup>6</sup>
Standard 2	24.8	1 x 10 <sup>5</sup>
Standard 3	28.4	1 x 10 <sup>4</sup>
Standard 4	32.6	1 x 10 <sup>3</sup>
Standard 5	36.1	1 x 10 <sup>2</sup>
Unknown sample	31.7	2.93 x 10 <sup>3</sup>
Negative control	40.2	-

## Indicator organisms – a reliable approach for general hygiene control

The presence of specific gastrointestinal microorganisms in water is frequently used as an indicator for fecal contamination. *Escherichia coli*, shiga toxin 1 and/or shiga toxin 2-producing *E. coli* (STEC) as well as shiga toxin-producing *Shigella dysenteriae* can get into water, e.g., via fecal contamination or fertilizers. As this bears the risk of causing severe dysentery, hemorrhagic colitis, and hemolytic uremic syndrome water quality evaluation regarding *E. coli* is obligatory\*\*. Several assays, including innuDETECT E. coli O157, are designed for the highly specific detection of such pathogens. Figure 2 and Table 2 show a sample application with the combination of three innuDETECT Assays for the characterization of EHEC O104:H4¹.



<sup>&</sup>lt;sup>1</sup> strain that caused the HUS epidemic in Germany 2011

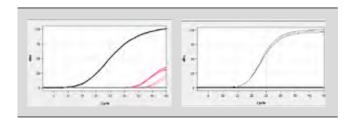


Figure 3/Table 3: Differential detection of *Legionella* in water samples obtained from cooling towers. *Legionella spp.* (left) are detected in the Cy5 channel and *Legionella pneumophila* (right) in the FAM channel.

	Legionella spp.	Legionella pneumophila	
Water sample 1	(weakly) positive	negative	
Water sample 2	negative	negative	
Water sample 3	positive	negative	
Positive control (PCR)	positive	positive	
Negative control (PCR)	negative	negative	

#### Advanced pathogen detection – Ruling out specific hazards

Clostridia spp. are bacteria generally found in all environmental habitats including water. Its detection is obligatory according to drinking water regulations\*\*\*. The detection via conservative cultivation entails several critical issues. That is why PCR-based assays offer an indispensable alternative. Moreover, Yersinia enterocolitica is widely present, especially in animal reservoirs and contaminations are observed in drinking and surface water posing a severe risk to health. Both innuDETECT Clostridium perfringens Assay and innuDETECT Yersinia enterocolitica Assay offer the perfect solution for fast and sensitive detection of the respective pathogens while the included internal controls ensure result reliability. This is clearly shown in Figure 4 and Table 4.

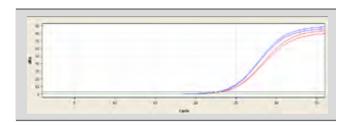


Figure 2/Table 2: Characterization of *E. coli* O104:H4 by real-time PCR via combination of innuDETECT Assays for E. coli O104 (red), Shiga Toxin 1 (green), and Shiga Toxin 2 (blue). Shown are signals detected in FAM channel and corresponding results (positive/negative).

innuDETECT Assay for	Signal FAM channel		
E. coli 0104	positive		
Shiga Toxin 1	negative		
Shiga Toxin 2	positive		

## Differential pathogen detection – Co-identification of most relevant subspecies

The presence of *Legionella* needs to be tested in drinking and process water, with a special focus on the subspecies *L. pneumophila*. Legal regulations such as the 42. BlmSchV require the evaluation of this Legionnaires' disease-causing pathogen and aims to prevent its emission from evaporation coolers and cooling towers. Identification of *Legionella* by conventional methods is highly challenging and hampered by co-presence of other microorganisms. The innuDETECT Legionella Assay overcomes these issues and allows for the simultaneous detection of total *Legionella spp.* and *L. pneumophila* according to the ISO/TS 12857 within a single reaction. Figure 3 and Table 3 show the results of *Legionella* detection and differentiation within water samples from industrial cooling towers. The presence of *L. pneumophila* could only be detected in the positive control of the PCR.

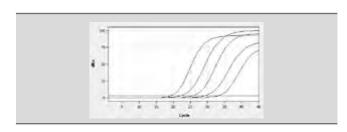


Figure 4/Table 4: Detection signals and ct values in FAM channel during real time PCR were obtained from *C. perfringens* DNA dilutions using the innuDETECT Clostridium perfringens Assay in FAM channel.

DNA amout	Ct value Clostridium perfringens (FAM)
1 ng	18.44
100 pg	21.69
10 pg	25.28
1 pg	28.99
100 fg	33.56
Negative control (PCR)	No ct

<sup>\*\*\*</sup> Deutsche Trinkwasserverordnung (2001) requires determination Clostridium perfringens according to DIN EN ISO 14189:2016-11

## **Order Information**

#### innuDETECT Assays

Order number (24 or 96 reactions)	Product name	Starting material	Assay time	qPCR detecion channels	Sensitivity
845-IDF-0031024 845-IDF-0031096	innuDETECT Bacteria Quantification Assay	Extracted bacterial DNA	~ 60 min	<ul><li>FAM (Bacteria)</li><li>HEX (Internal Control)</li></ul>	Dependent on background of negative control
845-IDF-0033024 845-IDF-0033096	innuDETECT Legionella Assay	Extracted bacterial DNA	~ 90 min	<ul> <li>FAM (Legionella pneumophila)</li> <li>Cy5 (Legionella spp.)</li> <li>HEX (Internal Control)</li> </ul>	<ul> <li>Legionella pneumophila: up to 10 DNA copies/PCR</li> <li>Legionella spp.: up to 20 DNA copies/PCR</li> </ul>
845-IDF-0034024 845-IDF-0034096	innuDETECT Clostridium perfringens Assay	Extracted bacterial DNA	~ 90 min	<ul><li>FAM (Clostridium perfringens)</li><li>HEX (Internal Control)</li></ul>	Up to 10 DNA copies/PCR
845-IDF-0032024 845-IDF-0032096	innuDETECT Yersinia enterocolitica Assay	Extracted bacterial DNA	~ 90 min	<ul><li>FAM (Yersinia enterocolitica)</li><li>HEX (Internal Control)</li></ul>	Up to 1 copy/PCR
845-IDF-0027024 845-IDF-0027096	innuDETECT E. coli O157 Assay	Extracted bacterial DNA	~ 60 min	<ul><li>FAM (E. coli O157)</li><li>HEX (Internal Control)</li></ul>	Up to 5 DNA copies/PCR
845-IDF-0028024 845-IDF-0028096	innuDETECT E. coli O104 Assay	Extracted bacterial DNA	~ 60 min	<ul><li>FAM (E. coli O104)</li><li>HEX (Internal Control)</li></ul>	Up to 5 DNA copies/PCR
845-IDF-0025024 845-IDF-0025096	innuDETECT Shiga Toxin 1 Assay	Extracted bacterial DNA	~ 60 min	<ul><li>FAM (Shiga toxin 1)</li><li>HEX (Internal Control)</li></ul>	Up to 5 DNA copies/PCR
845-IDF-0026024 845-IDF-0026096	innuDETECT Shiga Toxin 2 Assay	Extracted bacterial DNA	~ 60 min	<ul><li>FAM (Shiga toxin 2)</li><li>HEX (Internal Control)</li></ul>	Up to 5 DNA copies/PCR

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