Enterococcus faecalis
Quantitative DNA Standard for Water Testing

Enterococcus faecalis is a gram-positive cocci commonly isolated from the intestines of humans and animals. The presence of E. faecalis in water is one of the most widely accepted indicators of fecal pollution and the potential presence of enteric pathogens. For this reason, drinking & recreational water are routinely tested for the presence of Enterococci using E. faecalis as a control organism. EPA Method 1611 describes a qPCR-based method for faster sampling turn-around times when performing routine analysis of ambient marine and fresh recreational water quality.

EPA Method 1611 describes a procedure that measures the large subunit ribosomal RNA (lrRNA, 23S rRNA) target sequences from all known species of Enterococci in water. Purified, RNA-free, quantified, and characterized Enterococcus faecalis strain ATCC® 29212™ genomic DNA is recommended as the standard for this method. The Enterococcus faecalis Quantitative DNA Standard (ATCC® 29212Q-FZ™) was developed to meet the unique needs of this qPCR-based Method by providing a set of 3 DNA dilutions at known concentrations.

Kit Components
ATCC® 29212Q-FZ™ is provided as a kit comprised of three vials (100 µL each), containing dilutions 1, 2, and 3 which represent, respectively, 40,000, 4,000, and 400 Target Sequence Concentrations per 5 µL of frozen DNA standard. The Enterococcus faecalis Quantitative DNA Standard is useful for establishing calibration curves to measure the performance of assay platforms used in nucleic acid-based testing.
**Analysis of ATCC® 29212Q-FZ™**

DNA standard dilutions were analyzed on the Applied Biosystems® 7500 DX Real-Time PCR platform using TaqMan® Environmental Master Mix 2.0 (Life Technologies, Cat. No. 4396838); threshold was adjusted to 0.03. Copy numbers determined for the current lot of ATCC® 29212Q-FZ™ are shown in the figures provided. DNA copy number may vary depending on the quantitation method used.

<table>
<thead>
<tr>
<th>Dilution No.</th>
<th>TSC/Reaction</th>
<th>Ct Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>40000</td>
<td>21.138 ± 0.120</td>
</tr>
<tr>
<td>2</td>
<td>4000</td>
<td>24.328 ± 0.086</td>
</tr>
<tr>
<td>3</td>
<td>400</td>
<td>27.910 ± 0.110</td>
</tr>
<tr>
<td>4*</td>
<td>40</td>
<td>32.006 ± 0.282</td>
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</tbody>
</table>

*Please note that this product does not include Dilution 4 listed in the table and graph above. For all endusers following EPA water testing methods that specify the use of a dilution containing 40 TSC/5 µL, such as EPA Method 1611, it is recommended that this dilution be prepared at each enduser’s laboratory facility prior to use by diluting either Dilutions 1, 2 or 3 to achieve 40 TSC/5 µL. Long-term storage of dilutions at or below 40 TSC/5 µL is not advised due to the unstable nature of low-concentration preparations.

To learn more about water testing reference standards, please visit us online at www.atcc.org/waterQC.