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Case study - Wastewater

Quantitative PCR for Genetic Markers of Human Fecal Pollution.

Orin C. Shanks, Catherine A. Kelty, Mano Sivaganesan, Manju Varma, and Richard A. Haugland Appl and Envir. Microbiol. 2009. Vol 75.

Overview

- Keywords: Waterborne disease, environmental waters, microbial community, DNA extraction
- Aim of the study: Development of a method to assess microbial community present in waste water sample
- Application: Quantitative PCR
- Sample name: Wastewater
- Material: FastPrep-24™ instrument, FastDNA™ Spin Kit for Soil containing Lysing Matrix E
- Buffer: Sodium Phosphate Buffer and MT Buffer supplied with the FastDNA™ Spin Kit for Soil

Protocol and Parameters

500ml of primary effluent was collected and immediately stored on ice. 25 ml of each sample was filtered through a 0.2-µm-pore size supor-200 filters and each filter was placed in a sterile 1.5 ml microtube and stored at -80°C. For DNA extraction.

- 1. Cut the freezed filters with a sterile cutter.
- 2. Add the cutted filters to a Lysing Matrix E tubes.
- 3. Add 978 µl of Sodium Phosphate Buffer and 122 µl of MT buffer, provided with the FastDNA™ spin kit for Soil.
- 4. Homogenize in the FastPrep-24™ instrument for 120 seconds at a speed setting of 6.0
- **5.** Centrifuge at 14,000 x g for 5-10 minutes to pellet debris.
- 6. Proceed with the FastDNA™ Spin Kit for Soil extraction protocol.

Conclusion

- The FastPrep-24[™] and associated matrices have demonstrated **successful lysis and DNA extraction** from 20 samples of wastewater in only 120 seconds.
- This method saves hours of work during sample preparation, ensures high purified DNA and an effective PCR amplification.

Successful sample preparation using the MP Biomedicals FastPrep® product line has been highlighted in thousands of scientific articles. To access articles and other materials, visit www.mpbio.com/FastPrepLibrary.

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