

Determination of Bacteria Retention in the Thermo Scientific Barnstead GenPure Pro Water Purification System

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Key Words

Bacteria, Ultrapure Water, Thermo Scientific Barnstead, GenPure, ASTM Type 1, E.coli

Abstract

The bacteria retention of a Thermo Scientific™ Barnstead™ GenPure™ Pro water purification system was evaluated using the membrane filtration method based on the European Pharmacopoeia method described in Chapter 2.6.12.¹

Introduction

Bacteria are single-celled organisms that can be found nearly everywhere in a busy laboratory. Although many of these bacteria are harmless to a person's health, they can create unwelcome variability in an experiment. Because of their abundance and ability to be easily transferred, precautions such as aseptic technique are employed. Using bacteria-free water during steps such as sample preparation, system rinsing, or buffer preparation is an easy method of reducing the chance of bacterial contamination.

Water purification systems are a reliable source for bacteria-free water. On average, bacteria such as *Escherichia coli* (*E. coli*), which are widely used in the laboratory, have an length of about 2 μM, and an average diameter of 0.5 μM.² A 0.2 μM absolute membrane filter at the end of the system is used to remove any particles or bacteria that are larger than the pore size of the filter.³ Proper maintenance of the water system, including filter replacement as specified in the manual, helps to ensure the water remains bacteria-free.

Ultrapure water from a Barnstead GenPure water purification system was analyzed for the presence of aerobic bacteria. The GenPure Pro system was chosen from the family of GenPure systems, which also includes the GenPure and GenPure xCAD Plus models. All of these systems have the same feed water requirements, basic flow path and all dispense water through a 0.2 μM final filter. Because these systems require pretreated feed water, the feed water must be treated by reverse osmosis (RO) or deionization (DI) to meet the incoming feed water requirements listed in the system's operational manual⁴.

In normal operation, the acceptable incoming bacteria concentration for the system's feed water is defined as <100 CFU/ml. To challenge the system, the GenPure Pro system was connected to a tap water line rather than its normal pre-treated feed water, to help determine if the increased bio-load would impact the system's ability to retain the bacteria.

Materials and Methods

Bacteria determination of the GenPure Pro system using pre-treated feed water

As stated in the introduction, the GenPure Pro system is an ultrapure water system, which requires pre-treated water, otherwise known as a polisher system. To pre-treat the feed water for the GenPure Pro system, a Thermo Scientific™ Barnstead™ Pacific™ TII 20 UV water purification system was set up according to the operational manual⁵, along with a 60 L Thermo Scientific reservoir, as demonstrated in Figure 1. The system was put into operation and 70 L of water was dispensed from the storage reservoir and discarded. The GenPure Pro was then connected to the reservoir and rinsed with 10 L of water.

Water sampled from the 60 L storage tank, as well as the ultrapure water produced from the GenPure Pro system, were tested for bacterial growth using the membrane filtration technique¹. Clean techniques were used to reduce the chance of environmental contamination.

After dispensing 0.2 L of water from the reservoir and GenPure Pro system, 1 L samples were collected in sterile flasks at the outlet of the storage reservoir and the GenPure Pro system to probe the water qualities.

Figure 1. Diagram of a Pacific TII water system with a 60 L reservoir feeding a GenPure Pro water system.

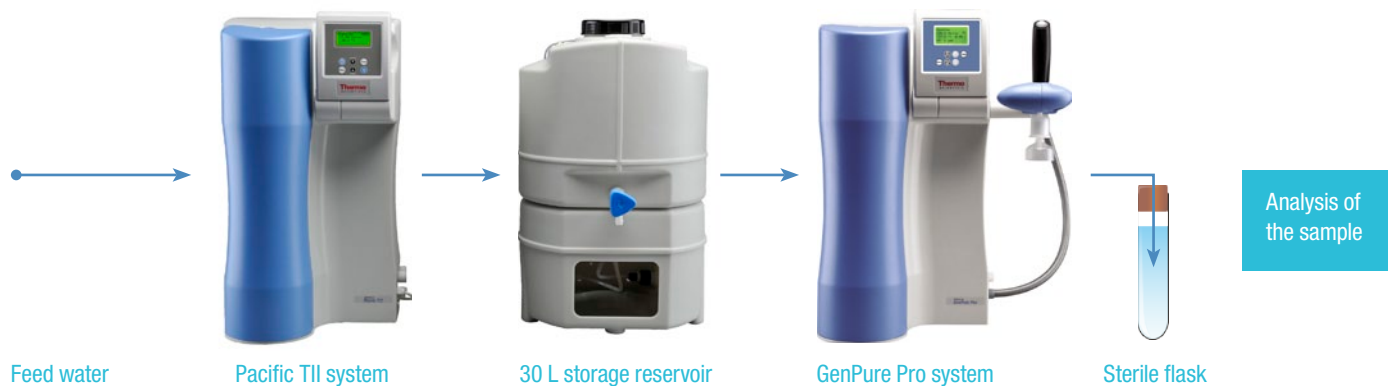


Figure 2. Diagram of the set-up for the GenPure Pro water system being fed tap water



Figure 3. Pictures of the CN membrane treated with tap water (left) and treated with ultrapure water produced from the GenPure Pro system fed with tap water (right)



The complete 1 L sample was filtered through a 0.2 μm cellulose nitrate (CN) membrane. The membrane was then transferred aseptically to a R2A-Agar and incubated at 35° C for 5 days in a Thermo Scientific™ Heratherm™ compact microbiological incubator (model IMC18). The entire procedure was repeated after all consumables in the systems and reservoirs were replaced with new ones to produce two sets of data.

Bacteria determination of the GenPure Pro system using tap feed water

The previously tested GenPure Pro system was disconnected from the Pacific TII system and 60 L reservoir and connected to tap water to create a new feed water source, as shown in Figure 2.

To equilibrate the system on tap water, 30 L of tap water was rinsed through the GenPure Pro system and then 1 L samples were collected at the inlet and outlet of the GenPure Pro system to probe the bacteria concentration for both the feed and product water. The 1 L sample was filtered through a 0.2 μm cellulose nitrate (CN) membrane, which was then transferred aseptically to a R2A-Agar and incubated at 35° C for 5 days in a Heratherm compact microbiological incubator (model IMC18). After 5 days, the colonies were counted for all 4 samples and the amount of bacteria per ml (CFU/ml) in the feed and product water was calculated. The entire procedure was repeated after all consumables in the systems were replaced with new ones to produce two sets of data, for a total of 8 samples.

Results

After 5 days, any bacteria colonies found on the R2A-Agar plates were counted (as shown in Figure 3) and the amount of bacteria per ml in the water were calculated. The data is summarized in Tables 1 and 2.

Conclusion

The GenPure Pro system was able to filter bacteria down to < 0.01 CFU/ml, even when challenged to purify tap water.

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Table 1: Bacteria count from the storage reservoir and the GenPure Pro system

	Storage Reservoir Water – Run 1	Storage Reservoir Water – Run 2	GenPure Water – Run 1	GenPure Water – Run 2
Bacteria CFU/ml	>10	> 10	<0.01	<0.01

Table 2: Bacteria count from the tap water and from the GenPure Pro system

	Tap Water – Run 1	Tap Water Water – Run 2	GenPure Water – Run 1	GenPure Water – Run 2
Bacteria CFU/ml	>10	> 10	<0.01	<0.01

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