Application Note

Thermo Scientific Nalgene Analytical Filters pass ISO 7704 requirements for water quality testing

Robert Scott, Matthew McCoy, and Joseph Granchelli

Key Words
Analytical Filter Units, water quality testing, cellulose nitrate membrane, microbial recovery

Introduction

Analytical filter units and filter funnels are often used in water quality testing to determine the microbial content of samples. Producing accurate results in this type of testing is dependent on the filter's performance in retaining microbes on the membrane. To be counted, microbes must be retained by the membrane; any microbial load not retained will not be reflected in the final results. In order to test this performance in Thermo Scientific™ Nalgene™ Analytical Filter Units and Filter Funnels, their functionality was assessed by microbiological recovery, as described in ISO 7704-1985 (E). This test evaluates the bacteriostatic and fungistatic properties of membrane filters, and product functionality, post-manufacturing. According to the standard, greater than 80% of the colony forming units must be retained by the filter when compared to control plates. For this study, several different types of Nalgene analytical filters were tested using *Escherichia coli* and *Saccharomyces cerevisiae* as representative bacterial and fungal organisms.

Experimental details

ISO 7704 Requirements

At least 5 replicate samples are required for each lot. A minimum of 200 colonies is needed for statistical comparison with ideally between 25-100 colonies per plate. Membranes producing counts ≥80% of the control plate counts are considered acceptable.

Sample Preparation

Twenty samples, ten per organism, were randomly selected for each of the nine filter units and filter funnels tested (see Table 1 for catalog number). All membrane filters in this study were cellulose nitrate (CN mixed ester) of either 0.45µm or 0.2µm pore size. Stock cultures of *E. coli* ATCC# 11775 and *S. cerevisiae* ATCC# 7754 were used as the challenge organisms in this study. All cultures were prepared and maintained for 18 to 24 hours at 30 ± 2°C. Prior to filtration, the density of each culture was estimated using a spectrophotometer at 530nm to determine the dilution scheme to obtain a target of 25-100 colony forming units (CFUs) per plate. Non-selective medium Plate Count Agar (PCA, Remel SMA #R454702) was used for growing *E. coli* on both control and test plates, while Sabourad Dextrose Agar (SDA, Remel #R454462) was used for *S. cerevisiae*. 14x-xxxx series filter funnels were tested installed on a three-outlet manifold, 130-xxxx series filter units incorporate a receiver chamber so do not require a manifold.

Filtration

One milliliter of the overnight *E. coli* culture was serially diluted 10-fold with sterile deionized water to a 10^{-7} dilution. The *S. cerevisiae* culture was handled similarly except it was diluted to 10^{-5}. Using a calibrated micropipette, 0.5mL (500µl) of well-mixed culture dilution was added to 30mL of sterile deionized water in

Thermo SCIENTIFIC

Part No.	E. coli ATCC # 11775 Percent Recovery (R)	S. cerevisiae ATCC # 7754 Percent Recovery (R)	Meets ISO 7704 Requirements (R ≥ 80%)
130-4020	81%	81%	Yes
130-4045	100%	91%	Yes
140-4045	89%	81%	Yes
145-0020	86%	86%	Yes
145-0045	89%	88%	Yes
145-2020	100%	100%	Yes
145-2045	100%	100%	Yes
147-0045	100%	85%	Yes
147-2045	100%	100%	Yes

Table 1. All filter units and filter funnels tested show greater than 80% recovery, passing the ISO 7704 standard for water quality testing.

each filter unit or funnel. Vacuum was then applied, and once drained completely two 30 mL rinses of sterile deionized water were filtered. The membrane was then aseptically rolled onto a PCA plate using sterile forceps, ensuring complete and even contact with the agar surface. Pour plate controls for each lot were prepared by pipetting 0.3mL of the culture dilutions into each of 5 polystyrene culture dishes, which were over-poured with molten PCA, stirred gently and allowed to solidify. All test and control plates were inverted and incubated at 30 ± 2°C for 18 to 24 hours.

Plate Counting

After incubation was complete, all colonies on membranes and control plates were counted using a hand tally counter and recorded. The arithmetic mean of CFUs on the 10 sample plates for each lot was calculated, and was then divided by the mean of the five control pour plates and multiplied by 100 to yield the percent recovery (Formula 1).

Formula 1. $R = m_m / m_c \times 100$

Where

R = percent recovery

m_m = mean of test membrane counts

m_c = mean of control pour plate counts

Results and discussion

The requirements set forth by ISO 7704 have all been met. All nine filter products tested in this study achieved at least 80% recovery (see Table 1) for the two organisms used: *Escherichia coli* and *Saccharomyces cerevisiae*.

Conclusion

Thermo Scientific Nalgene Analytical Filter Units and Filter Funnels with cellulose nitrate membranes pass the microbial recovery requirements set forth in ISO 7704. Their performance in this application makes them an excellent choice for use in water quality testing.

References

International Standard ISO 7704 - 1985 (E). Water Quality – Evaluation of membrane filters used for microbiological analyses. International Organization for Standardization. First Edition – 1985-03-15.

USP <1117> Microbiological Best Laboratory Practices. USP 35 or current edition. Official from May 1, 2012.

ANZ: Australia: 1300 735 292, New Zealand: 0800 933 966; **Asia:** China Toll-free: 800-810-5118 or 400-650-5118;
India: +91 22 6716 2200, India Toll-free: 1 800 22 8374; Japan: +81-3-5826-1616; Other Asian countries: 65 68729717
Europe: Austria: +43 1 801 40 0; Belgium: +32 2 482 30 30; Denmark: +45 4631 2000; France: +33 2 2803 2180;
Germany: +49 6184 90 6000, Germany Toll-free: 0800 1-536 376; Italy: +39 02 95059 554;
Netherlands: +31 76 571 4440; Nordic/Baltic countries: +358 9 329 10200; Russia/CIS: +7 (812) 703 42 15;
Spain/Portugal: +34 93 223 09 18; Switzerland: +41 44 454 12 22; UK/Ireland: +44 870 609 9203
North America: USA/Canada +1 585 586 8800; USA Toll-free: 800 625 4327
South America: USA sales support: +1 585 899 7198 **Countries not listed:** +49 6184 90 6000 or +33 2 2803 2000

Thermo SCIENTIFIC
Part of Thermo Fisher Scientific

APLSPANFILTISO 0613