Membrane Filter Technique

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The Membrane Filter (MF) Technique was introduced in the late 1950s as an alternative to the Most
Probable Number (MPN) procedure for microbiological analysis of water samples. The MF Technique
offers the advantage of isolating discrete colonies of bacteria, whereas the MPN procedure only indicates the presence or absence of an approximate number or organisms (indicated by turbidity in
test tubes).

The MF Technique was accepted by the U.S. EPA for microbiological testing of potable water in the 11th edition of *Standard Methods for the Examination of Water and Wastewater. In the 1978 publication, Microbiological Methods for Monitoring the Environment, the U.S. EPA stated that the MF Technique is* preferred for water testing because it permits analysis of larger samples in less time.

Comparative Benefits

The concentration of larger samples on a membrane filter is a key benefit of the technique over the MPN procedure as well as over Pour Plate and Spread Plate techniques.

Many industry and U.S. EPA test procedures require samples of 100 mL or more to be analyzed for the presence of bacteria. The Pour Plate technique is limited to a sample volume of 2 mL and the Spread Plate technique has a limit of 0.5 mL.

Using the MF Technique, a 100 mL sample is passed through a 47 mm membrane using a filter funnel and
vacuum system. Any organisms in the sample are concentrated on the surface of the membrane. The filter is then placed in a petri dish with nutrient medium. The passage of nutrients through the filter
facilitates the growth of organisms on the upper surface of the membrane. The discrete colonies that
form on the surface of the membrane can be easily transferred to confirmation media.

Uses of the MF Technique

Municipal water treatment plants monitor drinking, waste, and surface water for the presence of coliform bacteria by the MF Technique. The key organism monitored in water treatment facilities is *E. coli. The U.S.* EPA considers this organism the leading indicator of fecal contamination.

In addition to its use by government labs for monitoring drinking water, the MF Technique is also used for microbial monitoring in the pharmaceutical, cosmetics, electronics, and food and beverage industries.

The MF Technique is used in these industrial labs to monitor the presence of microorganisms in process waters and final product.

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The pharmaceutical and cosmetics industries typically focus on monitoring their process water for *Pseudomonas species. The electronics industry monitors for any and all microorganisms because they*
must keep their process water free from even the smallest organisms. Microbial monitoring in the food and beverage industry typically employs several types of techniques because of the variety of samples that are encountered. Beverage samples can typically be monitored for microorganisms by the MF Technique, but when solid samples cannot be liquefied, alternative methods must be used

Advantages of the MF Technique

- Permits testing of large sample volumes.
- Reduces preparation time as compared to many traditional methods.
- Allows isolation and enumeration of discrete colonies of bacteria.
- Provides presence or absence information within 24 hours.
- Effective and acceptable technique. Used to monitor drinking water in government laboratories.
- Useful for bacterial monitoring in the pharmaceutical, cosmetics, electronics, and food and beverage industries.
- Allows for removal of bacteriostatic or cidal agents that would not be removed in Pour Plate, Spread Plate, or MPN techniques.

Step-by-step Procedures

1. Collect the sample and make any necessary dilutions.

2. Select the appropriate nutrient or culture medium. Dispense the
broth into a sterile Petri dish, evenly saturating the absorbent pad.
3. Flame the forceps, and remove the membrane from the sterile
package
4. Place the membrane filter into the funnel assembly.
5. Flame the pouring lip of the sample container and pour the sample
into the funnel.
6. Turn on the vacuum and allow the sample to draw completely
through the filter.

7. Rinse funnel with sterile buffered water. Turn on vacuum and allow
the liquid to draw completely through the filter.
8. Flame the forceps and remove the membrane filter from the
funnel.
9. Place the membrane filter into the prepared Petri dish.
10. Incubate at the proper temperature and for the appropriate time
period.
11. Count the colonies under 10 - 15 X magnification.
12. Confirm the colonies and report the results.

Thank you