Ready-to-Reconstitute Media Bags

Applicable to:

NCM3202	Buffered Peptone Water (BPW) (ISO) (10 x 20L)
NCM3203	Buffered Peptone Water (BPW) (10 x 20L)
NCM3205	Half Fraser (Demi-Fraser) Broth (5 x 20L)
NCM3206	Less Plus Medium (5 x 20L)
NCM3207	Buffered Peptone Water (BPW) (HQ) (10 x 20L)

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Intended Use

Ready-to-Reconstitute media bags contain exactly the correct amount of dehydrated culture media to prepare 20L of enrichment broth. Ready-to-Reconstitute Media Bags are not intended for use in the diagnosis of disease or other conditions in humans.

Description

The dehydrated culture media contain within each bag has been sterilized via irradiation and needs reconstituting with sterile deionized/RO water following the preparation instructions given below. Aseptic technique must be observed during reconstitution and use.

Typical Formulation and Appearance

See associated Technical Data Sheet; NCM0015 for BPW (ISO), NCM0003 for BPW, NCM0001 for Half Fraser Broth, NCM0202 for Less Plus Broth and NCM0270 for BPW HQ.

Additional Materials Required

-NCM3200 Filter Unit
-NCM3201 Quick Connectors
-Sterile tubing
- Reverse Osmosis (RO) or deionized water source
- Peristaltic pump
- Dispensing system

Precaution

Refer to SDS

Dehydrated Culture Medium Preparation

1. Prepare a sterile Filter Unit (NCM3200), ensuring any kinks in the tubing are removed.
2. Open the outer foil pouch of the ready-to-reconstitute media bag using the tear notch (do not use scissors for risk of cutting the inner bag). Remove the media bag and attachments carefully, and unfold.
3. Aseptically remove the white cap from the white Filter Unit tubing connector and the black cap from the media bag tubing connector. Retain both caps aseptically for later use.
4. Attach the white Filter Unit connector to the black connector on the media bag tubing.
5. Prepare a tube, running from a 20 liter RO/deionized water source via a peristaltic pump. For best results, prime the tubing to remove excess air before connecting to the filter unit via the barb (push on connection). Prime the pump on a slow setting initially to allow time for adequate removal of trapped air (step 6).
6. Before activating the pump to commence filling, release the slide clamp on the media bag tubing and note:

   for optimum filter performance, it is recommended that the filter be held at a 45° angle (with screw valve to be in the uppermost position) thus allowing full expulsion of air from the device. To

prevent buildup of pressure upon commencement of filling, slowly turn the screw valve on the side of the capsule filter to release trapped air from the filter unit. Do NOT fully remove the screw.

N.B. Purging air and filling of filter unit with water happens rapidly. Ensure the screw is securely tightened after releasing the air to avoid water leaks.

The speed of the peristaltic pump may then be increased.

7. During filling, manipulate the bag to aid dissolution of powder. Tubing should be monitored for buildup of pressure which can be identified by the tubing between the peristaltic pump and filter becoming rigid. Ensure the water supply does not run dry during filling – any air pumped into the filter unit will cause a buildu p of pressure within the connected systems. Care should be taken not to fill the media bag with more than 20 liters of water.
8. Once the bag has been filled with 20 liter RO/deionized water, turn off the pump. To drain any remaining water from the tubing into the bag, it may be necessary to gently squeeze the bag to force air into the tubing, alternatively, the tubing may be elevated and squeezed. The slide clamp should then be applied. It is recommended that the slide clamp be applied near to the bag rather than connector end of the tubing. Check the integrity of the bag and ensure powder is fully dissolved. At this point, the connectors should be unscrewed and the bag cap can be reapplied or the bag connector can be immediately connected to the dispensing system (step 10).
9. Remove the filter unit and reattach the cap. The Filter Unit may be autoclaved and reused; record the usage of the filter and discard after 100 liters or 5 filled bags. If the flow rate is greatly reduced at any time, the filter should be discarded and replaced.
10. Attach a sterilized Quick Connector (NCM3201, supplied separately) and sterilized tubing set to the bag tubing via the bag connector.
11. Connect the new tubing to a dispensing system or peristaltic pump, open the slide clamp and begin dispensing the reconstituted, sterile culture media as required. At low volumes it may be necessary to manipulate the bag to enable full drainage. Care should be taken not to allow air to be taken into system as this will adversely affect accuracy of dispensing.
12. When dispensing is complete, disconnect bag tubing from dispensing tube. Retain the quick connector and cap and sterilize for future use.
13. If the media bag is not emptied and media is required to be stored, close the slide clamp and use the sterile (if asepsis has been maintained) cap to reseal the bag tubing. Once the media bag is emptied, dispose with normal laboratory waste.

Expiration

Refer to expiration date stamped on the bag container. The dehydrated medium should be discarded if not free flowing, or if the appearance has changed from the original color. Expiry applies to medium in its unopened bag container when stored as directed.

Limitation of the Procedure

Due to nutritional variation, some strains may be encountered that grow poorly or fail to grow on this medium.

Storage

Bag (as supplied): store in the dark at 10-25 °C

Bag (reconstituted): store in the dark at 10-25 °C for up to 5 days (providing asepsis is maintained)

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