Guidance Document for Ordering and Usage of Trans-Isolate (T-I)

MenAfriNet

Toolkit

This is a guidance document that assists laboratorians with ordering, transportation, and handling of T-I media.

TOOL CONTENTS:

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Guidance Document for Ordering and Usage of Trans-Isolate (T-I)

I. What is T-I medium?

Trans-Isolate (T-I) is a biphasic medium used for the growth, holding, and transport of bacterial meningitis pathogens such as *Neisseria meningitidis*, *Haemophilus influenzae*, and *Streptococcus pneumoniae*. The solid phase of T-I is an agar slant that contains activated charcoal, soluble starch, and agar, while the liquid phase consists of a soy broth and supplement B (Figure 1). These supplements and nutrients will promote and support the growth of meningitis pathogens in both phases.

Figure 1. T-1 bottle.
Source: U.S. Centers for Disease Control and Prevention

II. Procuring T-I media from the World Health Organization (WHO)

- **Estimate the quantity of T-I that needs to be ordered for the season:**
  - To estimate the needs for the upcoming season, the national reference laboratory (NRL) will need to average the total number of specimens received from across the country in the previous two years. A 10% contingency may be added to this number. Historical information on the number of specimens received per year at the NRL can also be found in the WHO weekly meningitis bulletin. The actual amount to be ordered will be the difference between the needs for the season and the current stock inventory.
  - Laboratories will need to get an accurate count of how much unexpired T-I media they have on inventory in order to determine how much T-I media they will need to order. It is important to exclude those media expiring before the end of the season.
- **Ordering T-I:**
  - The NRL will need to work with the Ministry of Health (MOH) to place the orders through the WHO Country Office.
  - The MOH will initiate the request with the WHO Country Office via email or letter.
  - The WHO Country Office will forward the request to WHO Inter-Regional Support Team West Africa (ISTWA). The laboratory point of contact will need to be included in all correspondences.
  - Once the order is processed, ISTWA will ship the media to the WHO Country Office, who will then transfer the order to the MOH.
  - In order to improve ISTWA's production forecasts for the next season, a questionnaire will accompany the shipment (Appendix I). Laboratory staff are strongly encouraged to submit their feedback, as this will allow ISTWA to improve future forecasts.

III. T-I handling and transport

Quality control (QC)

- Non-inoculated T-I should be stored at 4°C immediately upon arrival in the laboratory.
- Remove T-I bottles from the refrigerator at least 30 minutes prior to inoculating it with the cerebrospinal fluid (CSF) specimen and allow it to warm to room temperature.
- The NRL should perform quality control on all new shipments of T-I before use. For each new lot and shipment, the following should be observed and documented:

  All laboratories (NRL, peripheral)

  - *Visual inspection*: Upon receipt, NRL and peripheral laboratory staff should visually inspect all media for signs of contamination and physical defects. Examples of contamination include turbid liquid, color change, or growth of bacteria or mold on the slant; physical defects include cracked glass, broken seal, leaking, reduced volume, or absent liquid phase. If any of the above occur, the T-I bottle should be discarded and ISTWA should be contacted for a replacement.

  NRL and regional laboratories with culture capacity

  - *Sterility testing:* Incubate one T-I bottle vented and one non-vented (refer to T-I inoculation and venting procedure below) for 48 hours at 37°C.
    - » Using a sterile syringe, withdraw 50 µl of liquid phase of T-I and plate onto a chocolate agar plate.
    - » Incubate the plate at 37°C with 5% CO_2 for 48 hours. Any growth on the media is indicative of contamination.
  - *Growth promotion*: Using sterile technique, inoculate one T-I bottle with 100 µl of an inoculum containing 103 CFU/ml for each of the following pathogens: *N. meningitidis*, *H. influenzae* and *S. pneumoniae*. Incubate the plate at 37°C with 5% CO_2 for 48 hours and observe for growth of bacterial colonies.

T-I inoculation

- Prior to inoculation, disinfect the rubber stopper of the T-I bottle with 70% alcohol by first lifting the small metal cap on top of the T-I bottle. Do not completely remove the aluminum cover (Figure 2).

Figure 2. Disinfecting the rubber stopper of the T-1 bottle.
Source: U.S. Centers for Disease Control and Prevention

- Inoculate the T-I with 0.5 - 1.0 ml of CSF using 21G (0.88mm) sterile syringe through the rubber stopper of the lid.
- Invert the T-I bottle several times and incubate the inoculated T-I at 37°C.
- Immediately label the T-I bottle clearly with the date, name of the patient, and/or epidemiological identification (ID/EPID) Country Code–Region – District–Year–Number (CCC- RRR- DDD- YY-NNNN). Include any other necessary information.

Transport of T-I media

- If T-I media cannot be shipped within 24 hours, ventilate with a cotton plugged needle inserted through the rubber stopper without touching the media (Figure 3) and incubate at 37°C. Before transporting the T-I media, remove the venting needle, disinfect the rubber stopper. Then transport in triple packaging following guidance for transport of infectious biological material. Transport T-I at ambient temperature.

Figure 3. Ventilating T-1 media with a cotton plugged needle.
Source: U.S. Centers for Disease Control and Prevention

- Dispose of all inoculated T-I in the same manner as infectious bacterial cultures.
- A case report form should be shipped along with the T-I to the national/reference laboratory as soon as possible.

Appendix I.

T–I Media Questionnaire

Please answer all questions and email form back to the WHO-IST MDSC point of contact. Please do not leave any questions unanswered.

I. CURRENT STOCK

1. How many T-I media remain unused and in your possession? _____

- ☐ When do these T-I media expire? _____/_____/_____ (dd/mm/yyyy)

II. NEW ORDER

2. How many T-I media have been requested? _____

- ☐ What date was the order submitted? _____/_____/_____ (dd/mm/yyyy)
- ☐ When did the order arrive to the laboratory? _____/_____/_____ (dd/mm/yyyy)
- ☐ How many T-I media were received by the laboratory? _____
- ☐ What is the date of expiration of the T-I received? _____/_____/_____ (dd/mm/yyyy)

III. QUALITY CONTROL (QC)

3. Did you visually check all T-I media for contamination upon arrival? _____

4. What are the storage conditions of the T-I in your laboratory?

- ☐ Stored at +2°C to +8°C
- ☐ Stored at room temperature? What temperature? _____
- ☐ What precautions do you take before distributing the T-I to other laboratories? _____

5. Did you perform QC on the T-I?

- ☐ Yes; how many?: _____
- ☐ No

6. Were there any issues with the QC?

- ☐ No
- ☐ Yes; please describe: _____

IV. DISTRIBUTION TO LABORATORIES

7. How many T-I media do you keep at your laboratory? _____

8. How many T-I media were shared with or given to other laboratories? _____

Please provide the quantity and name of recipients: _____

9. Of those distributed to the laboratories, how many inoculated T-I did you receive back? _____

10. How are T-I distributed from the national reference laboratory to peripheral laboratories?

- ☐ In a cooler
- ☐ At room temperature
- ☐ By other means, please describe: _____

11. How are T-I transported from peripheral laboratories to National Reference Laboratory?

- ☐ In a cooler
- ☐ At room temperature
- ☐ By other means, please describe: _____