Strain Conditioning for International Transportation of *Vibrio cholerae* O1/O139

GLOBAL TASK FORCE ON CHOLERA CONTROL

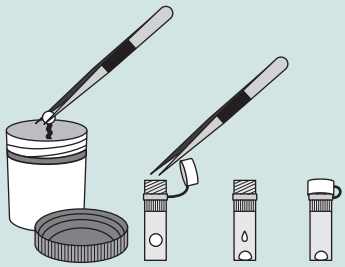
STRAIN CONDITIONING: 3 possible options [1]

Use gloves and lab coat when handling samples at all times.
Specimen Label: Carefully identify specimens. Indicate with permanent marker patient name, date of collection, time, location of sampling and location of patient when likely infected.
Lab Form: Use Annex 2B IDSR case-based laboratory reporting form[2].

CULTURE ON WET FILTER PAPER (WFP)

Use filter paper discs (6mm Ø, non-sterile), single-use forceps/needle, saline solution or broth medium, 2ml tube (screw cap).

Dip filter paper disk in a bacterial culture in broth medium (for example alkaline peptone water APW) with single-use device, transfer into tube, add 2 or 3 drops of saline or APW, close tube.

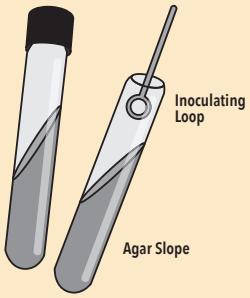


CULTURE INOCULATED ON NON-SELECTIVE MEDIUM

Use non-selective media (i.e. Mueller Hinton, brain heart infusion agar, trypticase soy agar).

Inoculate agar slant in tube.

Replace the cap after inoculation.



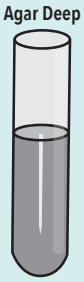
Incubate 18-24 hours at 37°C then cap tightly.

CULTURE INOCULATED ON STOCK CULTURE AGAR

Use semi-solid medium (non-selective) in tube.

Inoculation: insert a heavily loaded inoculating loop into the tube and push through the agar until it is approximately 1 cm from the bottom.

Replace the cap after inoculation.



Incubate 18-24 hours at 37°C then cap tightly.

CONSERVATION

No more than 2 weeks	Months	1 Year

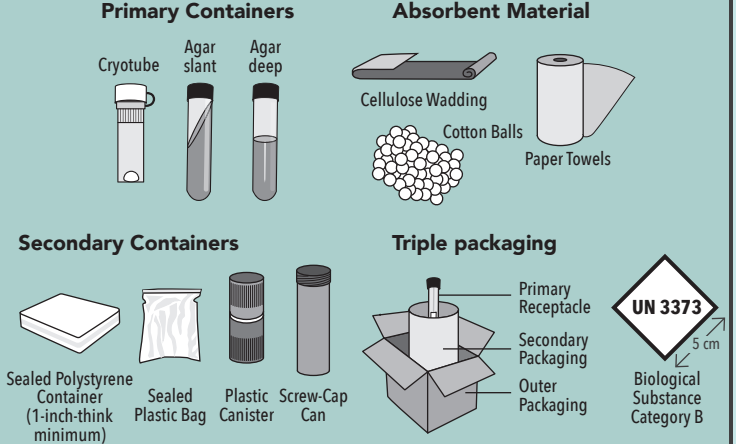
Ambient temperature (ideally 22-25°C). Do not refrigerate. Keep stool container out of direct sunlight.
Seal with tape or parafilm to prevent leakage (not required for dry filter paper).

INTERNATIONAL TRANSPORTATION (BY AIR, MUST REMAIN AT AMBIENT TEMPERATURE)

The shipper is responsible for ensuring the correct classification of infectious substances destined for transport and for the entire shipment until it reaches its final destination.

Sealed primary containers are individually wrapped, and absorbent material placed between the primary containers(s) and the secondary container. Both primary and secondary containers must be leak-proof.

The secondary container is then placed in a third container, usually a polystyrene or cardboard box, with appropriate padding interposed. The outer packaging must be rigid and have a minimum dimension of 100x100 mm.



Shipment by air must comply with local, national, and international regulations. For all media, follow IATA regulations for biological substances category B. There are no formal regulations for UN3373 packaging, however they must meet some requirements (see opposite panel).

Check-list: to be reviewed prior to shipment

✔ **Triple packaging (as shown), correctly marked and labelled:**
- Name, complete address and phone number of both sender and recipient
- Telephone number of a responsible person, knowledgeable about the shipment
- UN3373 labels (available at https://www.un3373.com/un3373-packaging/un3373/)
- Proper shipping name "Biological Substance Category B"

✔ **Required shipping documents:**
- Export license, local authorisation (if needed)
- Import permit (if needed)
- Lab request form indicating requested type of testing
- Customs/Proforma invoice, with a value for customs purposes of €10 (to be signed by the shipper)

IMPORTANT:
- Do not write the name of the organism on the outside of the package, only on the appropriate documentation in the package.
- Inform recipient laboratory about up-coming arrival of samples and provide any relevant shipping tracking details.
- Indicate storage and transport temperature requirements on the outer packaging.

GTFCC, April 2019

[1] Strains can also be frozen at -80°C in liquid nitrogen but it is not recommended for transportation because of the logistical complexity and high cost.

[2] Annex 2B - Case-based laboratory reporting form. https://www.afro.who.int/sites/default/files/2017-06/IDSR-Technical-Guidelines_Final_2010_0.pdf