Annex 8. Viral transport media (VTM)

a) Specimens from humans

WHO HQ in Geneva has stocks of commercially prepared viral transport media (COPAN Universal Transport Medium).

Another suitable commercially available medium is Eagle Minimum Essential Medium (E-MEM)

Alternatively, VTM can be prepared locally. A suitable VTM for use in collecting throat and nasal swabs from human patients is prepared as follows:

- Add 10g veal infusion broth and 2g bovine albumin fraction V to sterile distilled water (to 400 ml).
- Add 0.8 ml gentamicin sulfate solution (50 mg/ml) and 3.2 ml amphotericin B (250 µg/ml)
- Sterilize by filtration.

b) Specimens from animals

WHO recommends two different transport media for use when taking samples from animals. Of these the transport medium based on tissue culture medium 199 (A) is widely used for collection and transport of clinical specimens from all species. A second medium, the glycerol-based medium given below (B), provides longer-term stability of specimens where cooling is not immediately possible; it is suitable for egg inoculation but not suited for tissue culture inoculation.

Antibiotics and antifungals reduce the risk of bacterial and fungal contamination. With increasing use of these agents in animal husbandry it has become necessary to use high concentrations of them in transport media.

Antibiotics loose their effect over time if they kept at +4 °C and/or subjected to multiple freeze and thaw cycles. They should either be added to the transport medium when the samples are ready to be collected or added during preparation of the media which should then be frozen at -20 °C and thawed only when needed.

If attempts to isolate virus are to be made, transport media containing specimens should not be stored at -20°C but at -70°C or in a liquid nitrogen storage unit. Storage at -20 °C is adequate if the specimen is to be used for PCR tests only (See Section 4, Table 1).

A. Transport medium 199

1. Tissue culture medium 199 containing 0.5% bovine serum albumin (BSA)
2. To 1 litre of above add:
   - benzylpenicillin (2 x 10^6 IU/litre)
   - streptomycin (200 mg/litre)
   - polymyxin B (2 x 10^6 IU/litre)
   - gentamicin (250 mg/litre)
   - nystatin (0.5 x 10^6 IU/litre)

- ofloxacin hydrochloride (60 mg/litre), and
- sulfamethoxazole (0.2 g/litre)

3. Sterilize by filtration and distribute in 1.0–2.0 ml volumes in screw-capped tubes.

- OR -

B. PBS-Glycerol transport medium

1. Phosphate-buffered saline (PBS):

- NaCl 8g
- KCl 0.2g
- Na_2HPO_4 1.44g
- KH_2PO_4 0.24g
- Distilled water to make 1 litre

2. Autoclave PBS and mix 1:1 with sterile glycerol to make 1 litre

3. To 1 litre PBS/glycerol add:

- benzylpenicillin (2 x 10^6 IU/litre)
- streptomycin (200 mg/litre)
- polymyxin B (2 x 10^6 IU/litre)
- gentamicin (250 mg/litre)
- nystatin (0.5 x 10^6 IU/litre)
- ofloxacin hydrochloride (60 mg/litre), and
- sulfamethoxazole (0.2 g/litre)

Dispense 1.0–2.0 ml of transport medium into sterile plastic screw-cap vials (Cryovials). It is best to store these vials at –20 °C until used. However, they can be stored at +4 °C for 48–96 hours (optimally less than 48 hours) or at room temperature for short periods of 1–2 days.

Note:
Normal saline (NS) solution should **not** be used as a VTM. Adding BSA and antibiotics to NS changes the pH and this will destroy viruses.