



Title: *BPL-inactivation of aMPV*

No: RTLP-GL-VP-1

Location:
Old CCRC Tripp Lab

Approval Date:
10 September 2004

Supersedes Date:

Materials:

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|------------------------|---|----------------------------------|------------------|
| •Lab coat | •Phosphate-Buffered Saline (PBS) pH 7.4 | •Dialysis cassette (10,000 mwco) | •Pipettes |
| •Gloves | •Purified avian meta-pneumovirus (aMPV) | •dH ₂ O | •Pipetteman |
| •b-propiolactone (BPL) | •Vero Cells | •2L beaker | •Pipette Aid |
| •Stir plate | •4°C Cold Storage | •20mL beaker | •Pipetteman tips |
| •Stir bar | | | •25 gauge needle |
| | | | •5mL syringe |

Procedure:

1. Thaw 1mL of purified virus and place in small beaker with small stir bar. Add 1mL of sterile 1X PBS (pH 7.4). Add 0.01% BPL from -20°C freezer. Be extremely careful as BPL is very toxic.
2. Let stir at 4°C overnight. I usually place the stirrer in the cold room.
3. In the morning, chill down 1 liter of non-sterile 1X PBS. Wash an appropriate length of dialysis tubing (10,000 mwco) with distilled water several times. Clamp one end of the tubing with a dialysis clamp. Carefully add the BPL-virus to the tubing and clamp the other end.
4. Place the chilled PBS into a 2 liter beaker. Place a stir bar in the bottom of the beaker. Place the dialysis tubing in the beaker and return to 4°C. Stir gently at 4°C for 4 hours.

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5. After 4 hours, change out the PBS with fresh chilled PBS. Place the dialysis tubing into the PBS. Let stir gently overnight.
6. In the morning, pour off the PBS and add fresh PBS to the beaker. Let the tubing stay at 4°C stirring gently for 6 hours.
7. Use a 25 gauge needle to remove the BPL-inactivated virus from the dialysis tubing. Discard tubing and store the inactivated virus at -70°C.
8. Safety test 1:10 (or 1:20) dilution on Vero cells for 5-7 days.

Author	Management Approval/Date	Effective Date

