DNA Extraction from Baculovirus (for PCR)

- 1. Start with 100 µl of baculovirus stock (P0, P1, P2)
- 2. Add 100 µl of lysis buffer:
 - 10 mM Tris-HCl, pH 7.6
 - 10 mM EDTA
 - 0.25% SDS
- 3. Mix gently by pipetting
- 4. Add 100 µl of chloroform (1 volume)
- 5. Mix gently by inversion for 3–4 minutes
- 6. Centrifuge at 14,000 × g for 5 minutes
- 7. Carefully recover 150 µl of the aqueous phase (top layer)
- 8. Add 10 µl of 3 M sodium acetate, pH 5.2
- 9. Add 450 µl of 96% ethanol (3 volumes)
- 10. Mix gently and centrifuge at 14,000 × g for 15 minutes
- 11.Discard supernatant
- 12.Dry the pellet at 60 °C for 5 minutes
- 13. Resuspend DNA pellet in 20 μl of nuclease-free water
- 14.Use 1 μl of the prepared DNA in a 10 μl PCR reaction