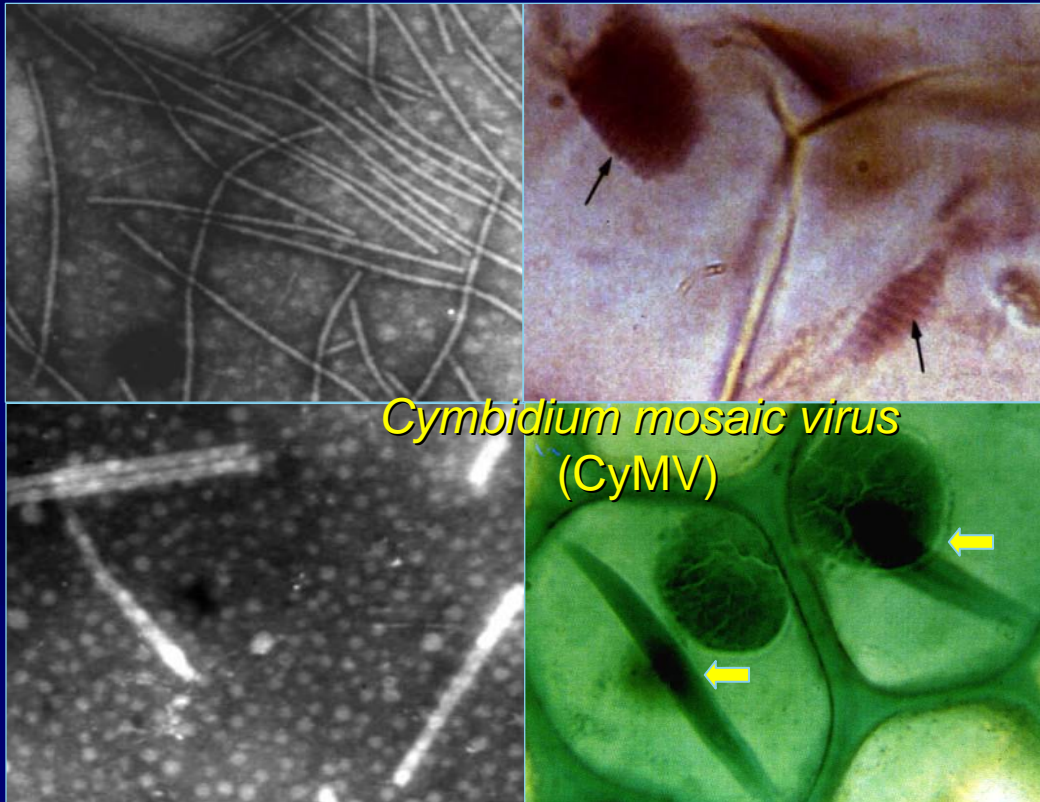


DIAGNOSIS OF CyMV AND ORSV ON *Dendrobium* AND *Phalaenopsis* THROUGH RT-PCR

Cymbidium mosaic virus (CyMV) and Odontoglossum ringspot virus (ORSV) belong to the potexvirus and tobamovirus groups. They are two of the most prevalent and economically deleterious orchid viruses, which infect numerous commercially important orchid genera, and have attained a world-wide distribution. CyMV induces floral and foliar necrosis while ORSV causes ring spots on leaves and color breaking on flowers. Mixed infections of both viruses can cause blossom brown necrotic streaks. The viruses also reduce plant vigor and lower flower quality, thus affecting their economic value. This subject applied RT-PCR method combining with the detection by agarose gel electrophoresis and checked result by real-time PCR. The result showed that these methods provide highly sensitive and specific tests for orchid virus diseases.

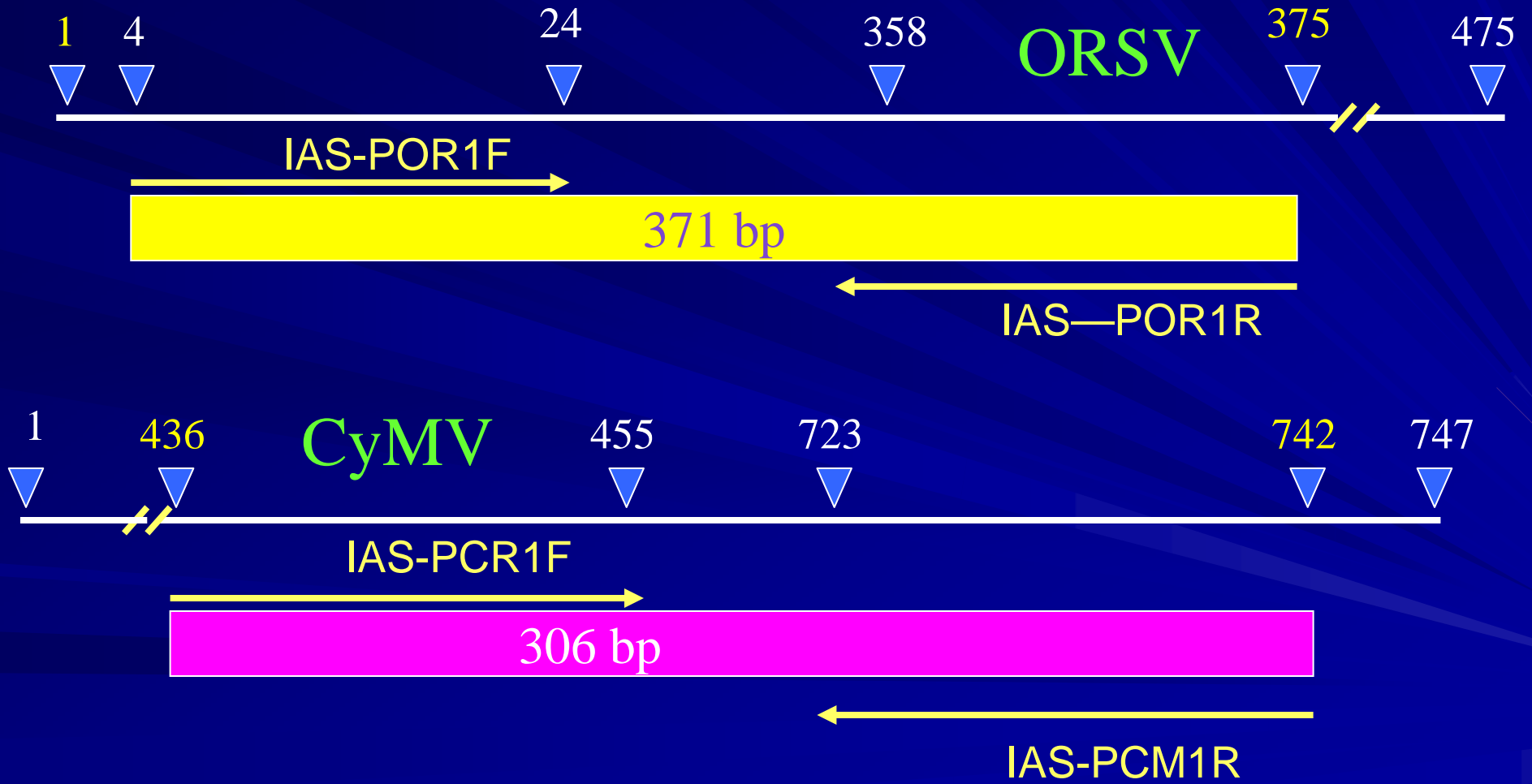
CyMV and ORSV in orchid observed through microscope



Cymbidium mosaic virus
(CyMV)

Odontoglossum ringspot virus
(ORSV)

Primer design for PCR and Mutiplex PCR



Primer to detect ORSV

IAS-POR1F: 5'-TTG TTC GAT TAC TAC AAT TAC-3' (21 nu)

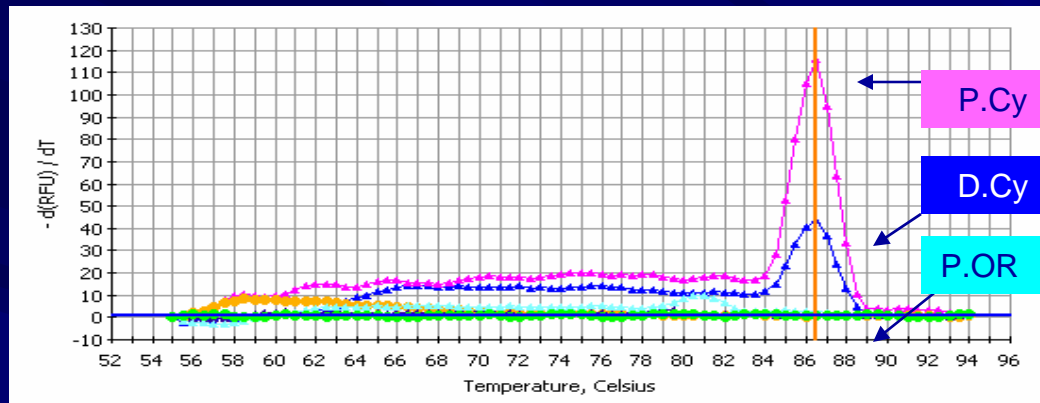
IAS-POR1R: 5'-TAA TAG AAC CAA ATG GGA-3' (18 nu)

Primer to detect CyMV:

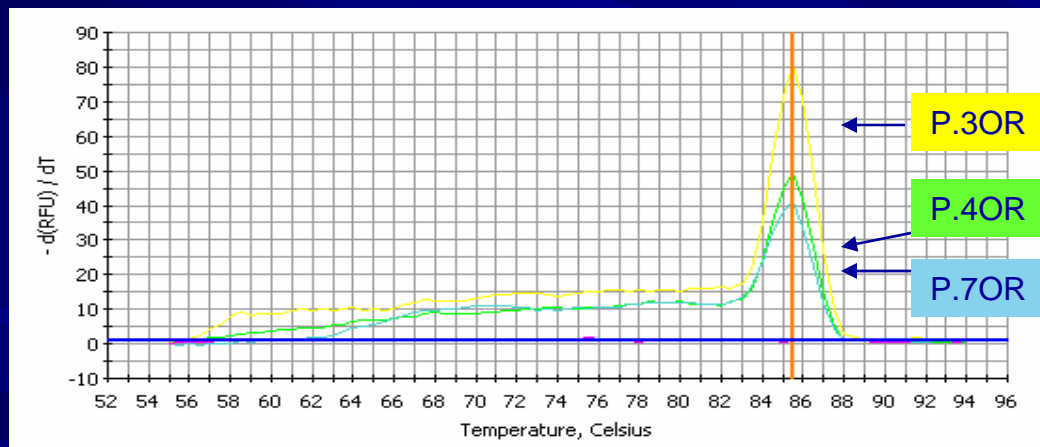
IAS-PCM1F: 5'-TAA AGA CTT ATT GCG TTA CG-3' (20 nu)

IAS-PCM1R: 5'-CAA CCA CTG CAG AGT GGA GT-3' (20 nu)

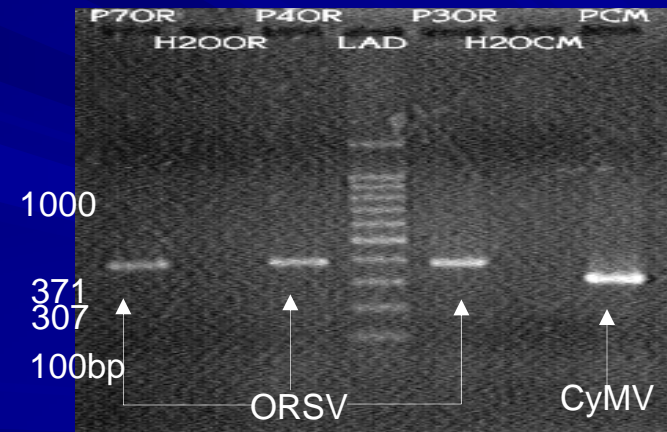
FastPCR was used to check primers



Melt Curve exhibited cDNA by CyMV virus

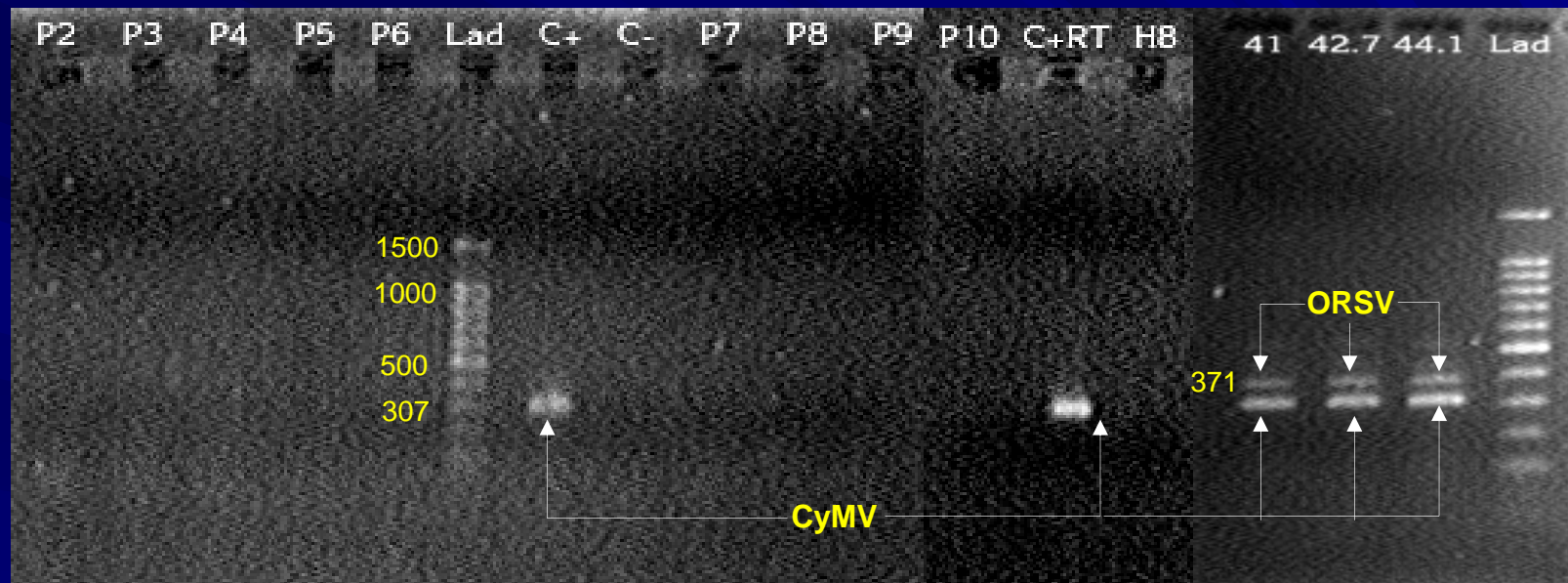


Melt Curve exhibited cDNA by ORSV virus

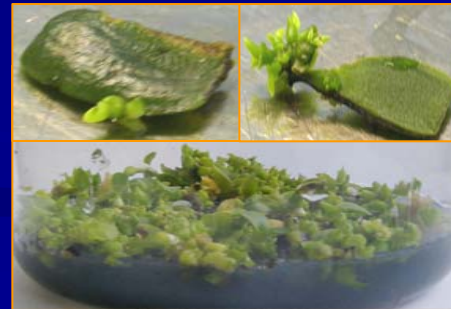


RT-PCR products on agarose gel for ORSV and CyMV

Multiplex PCR products on agarose gel to monitor virus in meristem and PLB to develop healthy plants



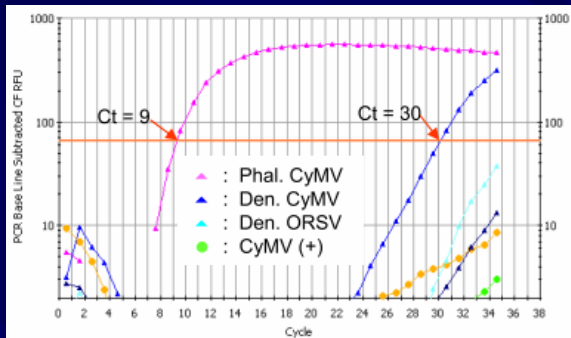
Shoot samples



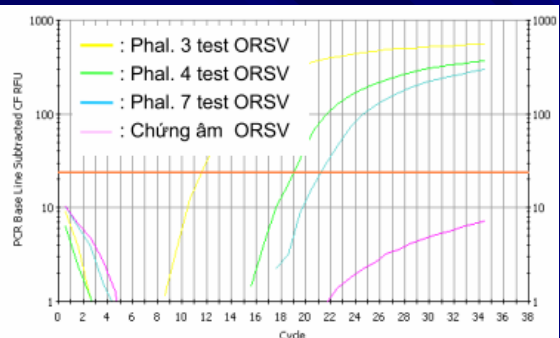
PLB samples (protocorm like body)

Table: Percentage of infected plants by CyMV and ORSV

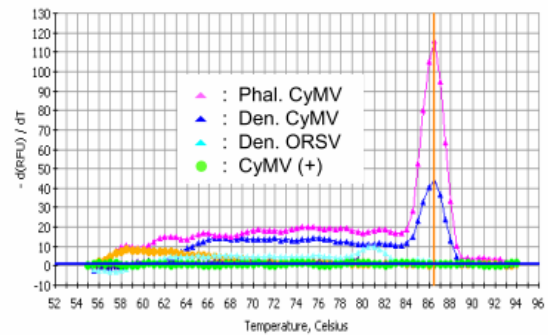
Collected sample	Sample number	Infected sample (%)	
		CyMV	ORSV
1 – Orchid farms	14	100	43
2 - <i>in vitro</i> orchids from laboratories	34	100	100
3 - <i>in vitro</i> orchids from seeds	12	0	0
4 - PLB regenerated from leaf tissues <i>in vitro</i>	34	0	0



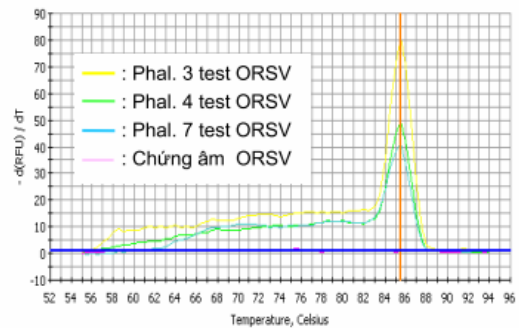
Hình 1: Mật độ huỳnh quang và số chu kỳ luân nhiệt



Hình 3: Mật độ huỳnh quang và số chu kỳ luân nhiệt



Hình 2: Biểu đồ Melt Curve



Hình 4: Biểu đồ Melt Curve