CHAPTER VI

SUMMARY

1. Isolation of total cytoplasmic RNA from rabies-infected BHK cells and canine brain specimen has been successful obtained using urea-SDS-phenol-chloroform. The technique is convenient, rapid and reproducible. RNA recovery is qualitative.

2. Reverse transcription-coupled PCR has been able to amplified rabies RNA template. Using PCR, specific portions of five structural genes of rabies virus have been cloned by specific primer designs. These amplified fragments were subcloned into *Sma*I site of pGEM-3Z. Recombinant plasmids has been named: pRAB-N, pRAB-NS, pRAB-M, pRAB-G and pRAB-L, possess insert portion derived from genes: N, NS, M, G and L, respectively.

3. Recombinant plasmids: pRAB-N, pRAB-NS, pRAB-M, pRAB-G and pRAB-L were served as rabies cDNA probes enabled to detect as low as 0.2-1.5 ng of rabies specific RNA by dot hybridization using ECL-random prime labelling and detection system.

4. Transcriptional events of rabies virus has been accomplished by PCR detection. Primary transcripts has been synthesized in the order of N, L, NS, M, and G, respectively. All rabies transcripts can be detected with in 5 hour postinfection. Data suggests that the primary transcription initiate at different promoter sites.

5. Dot hybridization detection of rabie virus RNA template has been revealed in canine brain RNA extracts using chemiluminescent-labelling system. Results are correlated to fluorescent-antibody test.