# CHAPTER VII

# CONCLUSION

In this study, we develop and evaluate the real-time PCR for diagnostic and serotyping dengue virus by using LightCycler system, hybridization probe format. Five oligonucleotide of hybridization were design to identify the melting temperature ($T_m$) of dengue virus genotype 1-4. First probe pair the detection probe labeled with LC-Red 640 was detected dengue genotype 1 and 3. The $T_m$ of this probe can discriminate DEN-1 and DEN-3. The second probe set were design to recognize the dengue genotype 2 and 4. Two anchor probes labeled with fluorescein in second probe set are perfect match with each genotype and the detection probe labeled with LC-Red 705 perfect match with genotype 4 and mismatch in genotype 2. Thus, the $T_m$ from this probe cannot discriminate the genotype 2 and 4. In addition, the in-house master mix can be used in the LightCycler system, which gives the similar result to the commercial kit. The sensitivity of real-time PCR are similar to nested PCR.

The applications of this study are useful in rapid diagnosis of dengue infection, using the less time-consuming, and use the in-house master mix instead of the commercial kit, which is decrease costs expenditure.