

## CHAPTER VI

### CONCLUSION

A total of 525 bacterial isolates was obtained from root nodules of seven soybean cultivars grown separately in soil samples from 15 subdistricts in Phisanulok province. Identical RAPD-PCR fingerprints using either RPO1 or CRL-7 as the primer of 105 isolates revealed they constituted 66 strains with 9 strains of fast-growers and 57 strains of slower-growers. All strains were found to contain *nifH* based on the annealing of RPO1 primer to their genomic DNAs. Multiplex PCR using the designed forward and reverse primers for *nodY* and *nodD1* genes could be used to detect the presence of fast- and of slow-growing soybean rhizobia as follows: fast-growing soybean rhizobial DNA yielded either 1300 bp or a combination of 500, 700, and 1500 bp while slow-growing soybean rhizobia yielded either 340 bp or 317 bp with 340 bp, or 317 bp with 340 bp and 657 bp or 317 bp with 657 bp, or 340 bp with 657 bp. The developed multiplex PCR was found to be specific for soybean rhizobia because when DNAs of *Agrobacterium tumefaciens* TISTR 507, *Xanthomonas campestris* TISTR 786 and *Proteus vulgaris* were used in the reactions, different or no PCR products were found. However, the developed multiplex PCR could not be used to distinguish certain groups of soybean rhizobia (fast-growing D11, D301, D384) and slow-growing soybean rhizobia (D291 and D481) because the 317 bp fragments formed in multiplex PCR reactions were found to have identical nucleotide sequence.