

Sirinrat Wannapinpong 2008: Cloning and Investigation of the Expression of *CHS-like* Gene Related to Curcuminoid Biosynthesis in Turmeric (*Curcuma longa* Linn.).
Master of Science (Genetics), Major Field: Genetics, Department of Genetics. Thesis
Advisor: Associate Professor Surin Peyachoknagul, D.Agr. 55 pages.

A part of *CHS-like* gene from *Curcuma longa* Linn. was amplified using nested PCR, which resulted in ~850 bp DNA fragment. This fragment was further ligated with pGEM®-T Easy vector. Each inserted part in the positive clones was cut with 7 restriction enzymes. Only 4 enzymes, i.e., *AluI*, *HinfI*, *MboI* and *RsaI*, showed polymorphic band patterns contributed to 6 different types. The nucleotide sequences of each type matched well with *CHS-like* genes from other plants. Southern blot hybridization indicated that the *CHS-like* gene contained at least 5 copies. The complete *CHS-like* gene, i.e., *ClCHS1*, *ClCHS2* and *ClCHS*, were cloned and their sequences were determined using TAIL-PCR. These genes were composed of 2 exons; the first exon encoded 64 amino acid while the second exon encoded 223-330 amino acid and having one intron of 82-95 base pair in between. The deduced amino acid sequences showed 50-60% homology to *CHS* and *CHS-like* genes from several other plants. Active sites, CoA binding sites, conserved sequences and substrate specificity of CHS superfamily enzymes were also found. Phylogenetic analysis based on amino acid sequences of *CHS*, *CHS-like* genes from 18 other plants indicated that the *ClCHS1*, *ClCHS2* and *ClCHS3* genes were belonged to the group of *CHS* and *CHS-like* gene from Angiosperm and formed separate group from others. Semi-quantitative RT-PCR analysis revealed that *CHS-like* was expressed at the highest level in the small rhizome size (<2 cm) and become less expressed in the longer ones.

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Thesis Advisor's signature

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