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Amorpha-4,11-diene Synthase of *Artemisia annua*: cDNA Isolation and Bacterial Expression of a Terpene Synthase Involved in Artemisinin Biosynthesis

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Artemisia annua, an indigenous plant to Korea, contains an antimalarial sesquiterpene, artemisinin. The first committed step of artemisinin biosynthesis is the cyclization of farnesyl diphosphate by a sesquiterpene synthase to produce an amorpha-type ring-system. The aims of this research were to molecularly clone and express amorpha-4,11-diene synthase for metabolic engineering. PCR amplification of genomic DNA with a pair of primers, designed from the conserved regions of sesquiterpene synthases of several plants, produced 184 bp DNA fragment. This fragment was used in the Northern blot analysis as a probe, showing approximately 2.2 kb of single band. Its sequence information was used to produce 2106 bp of a full-length cDNA sequence including 1641 bp of open reading frame for 546 amino acids (*kcs12*) through a rapid amplification of cDNA ends (RACE). The deduced amino acid sequence displayed 36% identity with 5-*epi*-aristolochene synthase of *Nicotiana tabacum*. Soluble fraction of *E. coli* harboring *kcs12* catalyzed the cyclization of farnesyl diphosphate to produce a sesquiterpene, which was identified through GC-MS analysis as amorpha-4,11-diene.