Abstract

Acetyl-CoA carboxylase (ACC) is a key enzyme of fatty acid metabolism with multiple isozymes often expressed in different eukaryotic cellular compartments.

In agriculture, inhibitors of plastid ACC are used as efficient herbicides against grass weed. However, grass weed populations resistant to aryloxyphenoxypropionate (APP) and cyclohexanedione (CHD) herbicides represent a major problem for sustainable agriculture. Using PCR and sequencing it was found out that five amino acid substitutions in plastid ACC were correlated with herbicide resistance of *Avena sterilis ssp. ludoviciana* Durieu populations from the northern grain-growing region of Australia: Trp-1,999-Cys, Trp-2,027-Cys, Ile-2,041-Asn, Asp-2,078-Gly and Gly-2,096-Ala. We showed, using a yeast gene-replacement system, that these single-site mutations also confer herbicide resistance to wheat plastid ACCase: Asp-2,078-Gly confers resistance to APPs and CHDs, Trp-2,027-Cys and Ile-2,041-Asn confer resistance to APPs, and Trp-1,999-Cys confers resistance only to fenoxaprop. These mutations are very likely to confer resistance to any grass weed species under selection imposed by the extensive agricultural use of the herbicides.

ACC provides an important target for new drugs to treat human diseases. We have developed an inexpensive nonradioactive high-throughput screening system to identify new ACC inhibitors. The screen uses yeast gene-replacement strains depending for growth on cloned human ACC1 and ACC2. In “proof of concept” experiments, growth of such strains was inhibited by compounds known to target human ACCs. Chemical libraries yielded new specific inhibitors of human ACC2. The target of the best of these inhibitors was confirmed with *in vitro* enzymatic assays. This compound inhibits human ACC2 with 2.8 μM IC_{50} and has no effect on human ACC1 at 100 μM.

We used the DNA sequence of wheat ACC also to study evolution. The DNA sequences of wheat ACC1 and ACC2 loci, encoding the plastid and cytosolic forms of the enzyme acetyl-CoA carboxylase, were analyzed with a view to understanding the evolution of these genes and the origin of the three genomes in modern hexaploid wheat. ACC1 and ACC2 loci from each of the wheats *Triticum urartu* (A genome), *Aegilops tauschii* (D genome), *Triticum turgidum* (AB genome), and *Triticum aestivum* (ABD genome), as well as two ACC2-related pseudogenes from *T. urartu* were sequenced. The 2.3– 2.4 Mya divergence time calculated here for the three homoeologous chromosomes, on the basis of coding and intron sequences of the ACC1 genes, is at the low end of other estimates. Our clock was calibrated by using 60 Mya for the divergence between wheat and maize. On the same time scale, wheat and barley

diverged 11.6 Mya, based on sequences of ACC and other genes. The regions flanking the ACC genes are not conserved among the A, B, and D genomes. They are conserved when comparing homoeologous genomes of diploid, tetraploid, and hexaploid wheats. Substitution rates in intergenic regions vary substantially along the loci and on average are 3.5-fold higher than the ACC intron substitution rates.