Genetic transformation and metabolic engineering of poppy

Sang-Un Park

Department of Biological Sciences, University of Calgary, Calgary,

Alberta T2N 1N4, Canada.

Introduction

Alkaloids are pharmacologically active, nitrogen-containing compounds produced predominantly, although not exclusively, in higher plants. The benzylisoquinoline alkaloids are the largest group in the field of plant alkaloids in terms of structural diversity and are found primarily in plants that belong to five families: the Berberidaceae, Fumariaceae, Menispermaceae, Papaveraceae, and Ranunculaceae. A variety of benzylisoquinoline alkaloids that remain important as pharmaceuticals are still isolated from plants, because of the complexity of their chemical structures.

The functions of benzylisoquinoline alkaloids in plants may include self-defense against herbivory and infection by pathogens. The pharmacological activity of benzylisoquinoline alkaloids that renders them useful as pharmaceuticals is often a clue to their biological role in the plant. For example, the effectiveness of morphine as an analgesic, colchicine as a microtubule disrupter, and (+)-tubocurarine as a neuromuscular blocker suggests that these alkaloids function as animal feeding deterrents. The antibiotic nature of sanguinarine suggests that it' s constitutive or inducible accumulation confers protection to the plant against challenges imposed by a pathogen. Many plants, such as opium poppy and California poppy, *Eschscholzia californica Cham* producing benzylisoquinoline alkaloids, invest considerable amounts of nitrogen and metabolic energy in the biosynthesis of numerous and structurally diverse alkaloids suggesting that these natural products play additional and essential ecochemical and/or physiological roles that remain to be discovered .

All benzylisoquinoline alkaloids share a common biosynthetic origin, beginning with the condensation of two aromatic units both derived from the amino acid L-tyrosine.

The enzymatic steps of particular interest in this proposal are: tyrosine decarboxylase (TYDC) which represents an entry point into the alkaloid pathway; *N*-methylcoclaurine hydroxylase (NMCH) which is the first of many P450-dependent enzymes in the early alkaloid pathway;

NADPH: cytochrome c reductase (CCR) which donates electrons to P450-dependent enzymes and is essential for their function; and berberine bridge enzyme (BBE) which operates at an important branch-point in sanguinarine biosynthesis. However, despite our extensive appreciation for the chemistry and enzymology of benzylisoquinoline alkaloid biosynthesis, the control architecture that regulates metabolic flux through the numerous biosynthetic pathways is only beginning to be understood. The application of molecular techniques to the study of benzylisoquinoline alkaloid biosynthesis will expand the frontiers of our ability to understand and manipulate these pathways in plants. The prospect to engineer the benzylisoquinoline alkaloid metabolism of plants for the 'custom' biosynthesis of pharmaceuticals will require both a thorough knowledge of the regulation of biosynthetic enzymes and genes, and the availability of cloned genes for the genetic transformation of plants and other organisms.