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Benzophenanthridine alkaloids, a subclass of the benzylisoquinolines, are produced in an *Eschscholtzia californica* (California poppy, used as a sedative by Native Americans). These alkaloids concentration increased with carbon source content in culture medium, suggesting that carbon source concentration is important factor for biosynthesis of benzophenanthridine alkaloids. We have performed proteomic analysis of proteins sequentially extracted from *E. californica* suspended cell which was cultured with various carbon source concentrations and separated by two-dimensional electrophoresis (2–DE). For the soluble proteins in the whole cell, the 2–D separation showed around 2,500 spots, detected under the conditions used in our experiments. And, we found 29 spots of which expression varied analogously with dihydrosanguinarine production. These results demonstrate the use of metabolite analysis as a tool for detecting target proteins related to metabolites production pathway. This study describes the first approach of soluble proteins profiling of *E. californica* cell culture.