Regiospecific Methylation of Flaonoid with O-methyltransferase from Bacillus cereus

Lee Hyo Jung, Kim Bong Gyu, Ahn Joong-Hoon*

Bio/Molecular Informatics Center, Department of Molecular Biotechnology, KonKuk University, Seoul

TEL : +82-2-450-3764, FAX +82-2-446-9001

Abstract

Regiospecific modificationhas been drawn attentions since it is difficult to be achieved with chemical synthesis. Biological reaction displayed high specific regiospecificity. Myriad of genes have been deposited in database, which might have a role in modification of natural compounds[1]. Flavonoids are one of the natural products produced from plants and have some potential functional groups that could undergo methylation or glycosylation[2]. We cloned a putative *O*-methyltransferase from *Bacillus cereus*, BcOMT2. It has 668 bp open reading frame which encodes 24.6kDa protein. In order to investigate the modification reaction mediated by BcOMT2, it was expressed in *E. coli* as His-tag fusion protein and purified with homogeneity. Several substrates such as caffeic acid, catechin, daidzein, naringenin, kaempferol, genistein, luteolin and quercetin were tested and reaction products were analyzed by thin layer chromatography (TLC) and high performance liquid chromatography (HPLC). BcOMT2 could transfer a methylation group to substrates that have a 3'functional hydroxyl group such as luteolin and quercetin. Comparison of HPLC retention time, UV/visible spectrum, and NMR data of luteolin and quercetin reaction product with authentic corresponding 3'-methylated product showed that the methylation position is at 3'-hydroxy group. Thus, BcOMT2 transfers a methyl group specifically to 3'-hydroxy group of flavonoids.

References

1. Ivanova,N., Sorokin,A., Anderson,I., Galleron,N., Candelon,B., Kapatral,V., Bhattacharyya,A., Reznik,G., Mikhailova,N., Lapidus,A., Chu,L., Mazur,M., Goltsman,E., Larsen,N., D'Souza,M., Walunas,T., Grechkin,Y., Pusch,G., Haselkorn,R., Fonstein,M., Ehrlich,D.S.D., Overbeek,R. and Kyrpides,N. Genome sequence of *Bacillus cereus* and comparative analysis with *Bacillus anthracis*. Nature 423, 87-91 (2003).
2. Joshi, C. P., Chiang, V. L., 1998. Conserved sequence motifs in plant *S*-adensyl-L-methionine-dependent methyltransferases. Plant Mol. Biol. 37, 663-674.