

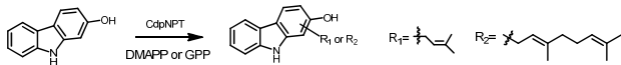
30E11-am08S

糸状菌由来インドールプレニル転移酵素を用いた非天然型新規プレニルカルバゾールの創出

○陳 静^{1,2}, 森田 洋行^{1,3}, 脇本 敏幸^{1,3}, 野口 博司², 阿部 郁朗^{1,3} (1東大院薬, 2静岡県大薬, 3JST-CREST)

【目的】 CdpNPT from *Aspergillus fumigatus* is an indole prenyltransferase (PT) that catalyzes a C5 prenylation of tryptophan-containing cyclic dipeptides, at N-1 of the indole core in the presence of DMAPP. Previously, we demonstrated *in vitro* combinatorial biosynthesis of unnatural novel prenylated indole polyphenols using a polyketide synthase (PKS1) and CdpNPT. Now we present precursor-directed biosynthesis of unnatural novel prenylated carbazoles by CdpNPT, and the biological activities of the obtained alkaloids. Moreover, to clarify the intimate structural details of the CdpNPT-catalyzed enzyme reactions, we also carried out crystallization study of CdpNPT.

【方法・結果】 CdpNPT was expressed in *E.coli*, as his-tag fusion protein at C-terminus, and was incubated with carbazoles in the presence of DMAPP (or GPP) in Tris-HCl buffer (pH 7.5) at 30°C for 16 hours. The products were analyzed by LC-MS and NMR spectra. The precursor-directed biosynthesis studies revealed that when 2-hydroxycarbazole was used as a prenyl acceptor, CdpNPT efficiently afforded not only two unnatural novel C5 prenylated carbazoles and one C5 diprenylated carbazole, but also two unnatural novel C10 prenylated carbazoles by using GPP instead of DMAPP. These are the first demonstration for the enzymatic prenylation of the carbazole scaffold by indole PTs. Further, we also succeeded in crystallization of CdpNPT. Crystallographic analyses of the enzyme are now in progress.



【文献】 Chen, *et al.*, manuscript in preparation (2011).